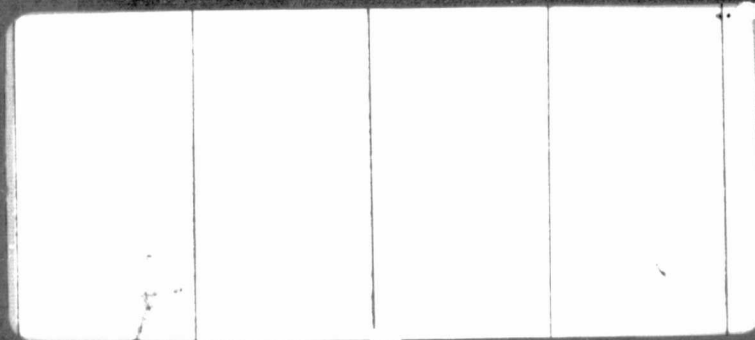


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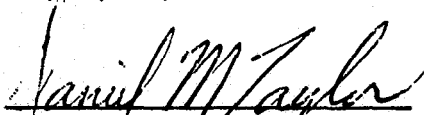
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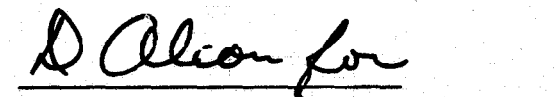
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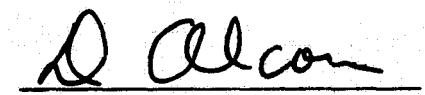
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PREFACE

This document contains a report on Research and Advanced Development at the Jet Propulsion Laboratory during the period 1 January 1975 to 30 June 1975, sponsored by the Planetary Quarantine branch of the NASA Office of Space Sciences and Applications.

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SECTION I
PLANETARY QUARANTINE STRATEGIES
FOR
ADVANCED MISSIONS
(NASA NO. 193-58-61-01)

ContentsTitle and Related Personnel

Subtask A
para. 1.1

STRATEGIES FOR SATELLITE ENCOUNTER

Cognizance: C. Gonzalez

Associate
Personnel: W. Brady

Subtask B
para. 1.2

OUTER PLANET ENTRY ANALYSIS

Cognizance: C. Gonzalez

Associate
Personnel: W. Jaworski

1.1 STRATEGIES FOR SATELLITE ENCOUNTER

1.1.1 Subtask A Introduction

The objectives of this task are to determine the impact of satisfying satellite quarantine on current outer planet mission and spacecraft designs; and to develop tools required to perform trajectory and navigation analyses for determining satellite impact probabilities.

1.1.2 Significant Accomplishments

The dominant planetary and satellite quarantine problem for the Jupiter orbiter mission is spacecraft reliability. The reason for this is there are a large number of potential spacecraft encounters with the Galilean satellites. If the spacecraft is in a near equatorial orbit, there are as many as eight (8) crossings of satellite orbit radii in one spacecraft orbit. If the period of satellite quarantine is 50 years, the probability of impact for an uncontrolled spacecraft is in the range of 0.1 to 1.0 (see Reference). Because of this, the overriding concern is not the probability of impact for a single encounter but what is the long term probability of impact if the spacecraft fails. The combination of the spacecraft reliability, the probability of contamination given impact, and the quarantine limit on probability of contamination will determine the severity of the problem. Unless some fail safe device is available, the Jupiter orbiter mission may have significantly reduced science yield due to planetary and satellite quarantine as a result of being forced to utilize inclined orbits.

In order to assess the severity of the reliability problem, the Mariner Jupiter Saturn (MJS) spacecraft reliability model was used to evaluate the probability of making a maneuver as a function of time from launch and time from the last maneuver. Some representative values are given in Fig. 1-A.1. Since Jupiter encounter occurs at about 800 days from launch, the probability of making a successful maneuver is about 0.99 at the beginning of the orbital phase if the maneuvers are separated by 10 days. The probability of success is reduced further if the maneuvers are separated by a larger time. Since there will probably be 3 or 4 maneuvers per orbit, a mission could not have very many orbits and satisfy the quarantine constraints as long as the long term probability of impact is high.

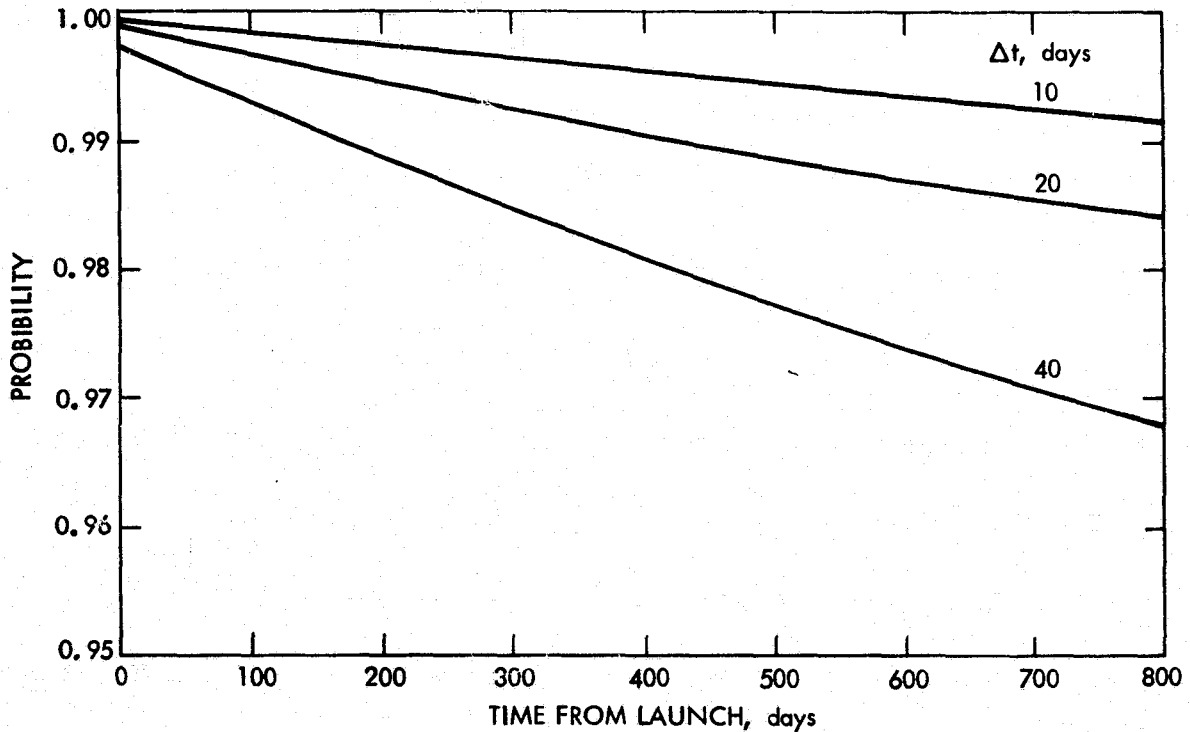


Fig. 1-A.1. Conditional probability of making the next maneuver

Figure 1-A.2 shows the relationship between collision likelihood and orbit inclination derived in the Reference . There is a dramatic decrease in the impact likelihood as the inclination is increased. The number of close encounters is also shown as a function of inclination and the percentage decrease is comparable. The disadvantage of the higher inclinations is the reduced potential science return. The near equatorial orbits provide a large number of close encounter possibilities which yield immediate science return plus the opportunity to use gravity assist maneuvers to set up future close encounters. As the inclination increases, these opportunities are reduced in frequency and as a result a longer mission is required to achieve a given set of science goals. The effect these conflicting factors have on the satisfaction of the quarantine constraints cannot be evaluated without further study.

Analysis software was developed to study the effect that the quarantine constraints might have on the maneuver strategy used in the orbiter mission. The maneuver strategies that were investigated did not provide adequate orbital

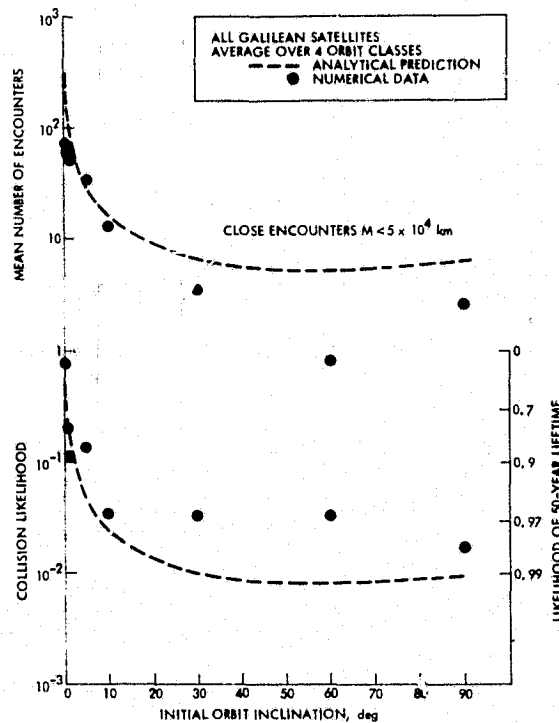


Fig. 1-A.2. Likelihood of close encounters and collision with the galilean satellites for a 50-year Jupiter Orbiter lifetime

control and, as a result, the relationship between quarantine and the maneuver strategy could not be evaluated. Because of the long term impact problem, it appears that quarantine will have little if any influence on the maneuver strategy.

Some further work is required to determine whether there is a means of providing some fail safe device so that the spacecraft reliability does not have such a dominant influence on the mission. Some consideration might be given to reducing the bioload on the spacecraft. The mission may be more cost effective with a cleaner spacecraft because of the increased science yield possible. The spacecraft reliability should also be given further consideration.

1.1.3 Future Activities

No future activity is planned.

1.1.4 Presentations

None

1.1.5 Reference

Friedlander, Alan L., "Jupiter Orbit Lifetime: The Hazard of Galilean Satellite Collision," Report Number SAI 1-120-194-T1, Science Applications Inc., Rolling Meadows, Illinois, Feb. 1975.

1.2 OUTER PLANET ENTRY ANALYSIS

1.2.1 Subtask B Introduction

The objectives of the outer planet atmospheric entry analysis subtask were to develop and use the tools to determine the thermal response characteristics of a typical spacecraft and related debris upon entry into the atmospheres of Jupiter, Saturn, Uranus and Titan.

Analyses have been performed to determine the aerodynamic and thermal responses of a spacecraft inadvertently entering the atmosphere of one of the outer planets, Jupiter, Saturn and Uranus or into Titan. (See references 1 through 5.) These analyses included calculations to determine the aerodynamic response of a spacecraft, major components occurring after breakup, and debris, and subsequent heating and thermal response of the materials. Some tests were also performed to determine the thermal response of some of the materials and components involved. A brief study was done of the situation with Uranus which indicated that if similar entry heating analyses had been performed results obtained in a Uranus study would probably be more analogous to Saturn results than those for Jupiter or Titan.

Analyses for Jupiter and Saturn were performed for each of three atmospheric models, (Titan two models), several entry angles, and component orientation in the atmosphere. This allowed comparison of results for a range of possible entry situations.

A spacecraft entering the atmosphere of an outer planet will begin to break up very early, i. e., by the time it reaches the continuum. During this process the heating will sterilize the exterior and lightweight components such as the thermal blankets; and the large spacecraft antenna will be completely sterilized. All appendages will also disintegrate. However, pieces of the bus or the ultraviolet spectrometer which have a lot of plastic material such as circuit boards and cabling will not be heated thoroughly enough to kill all organisms. The exact degree of heating received is a function of the entry conditions and the entry configuration of the component in question. Due to the difficulty in quantifying these factors, a conservative approach was used in determining microbial survival probability.

Therefore, in the case of Jupiter the most realistic and reasonably conservative approach is that a reduction in the encapsulation burden count due to entry heating of an order of magnitude (1 log reduction) can be safely assumed. The reduction in the exposed and mated burden of four orders of magnitude and three orders of magnitude, respectively, assumed for current outer planet missions still appears to be reasonable.

In addition to the aerodynamic and thermal analysis performed for large components, analyses were performed for particulate debris. The particulate debris is released in several ways. Debris is released early, either prior to entry as the spacecraft traverses interplanetary space and/or during early entry due to contact with the atmosphere. Previous analyses (see reference 1) indicate that the first mode of release did not lead to a significant number of particles reaching the planet. The second case has not been studied. Another release mode is due to spacecraft breakup upon entry into the atmosphere.

The small particle analyses have shown the existence of survival corridors for particles liberated early in the entry. However, very early in the continuum a point is reached in the atmosphere such that any debris liberated after that point will not contain viable organisms. This means that since in the case of Jupiter and Saturn, breakup takes place very early in the continuum, most debris will be sterilized. There is a change for some debris liberated right at, or slightly before the continuum regime, to contain some surviving organisms. Titan will have the same type of effect with only early released debris containing surviving organisms.

1.2.2 Significant Accomplishments

1.2.2.1 Analyses for Saturn and Titan. The current reporting period was concerned with large component analyses for entry into Saturn and Titan. In the case of Saturn, analyses were performed for those components in situations which either did not survive or for which survival was marginal in terms of organisms, in the case of Jupiter. Components which survived in the case of Jupiter would survive for Saturn also. Three components in particular were considered; thermal blankets, the magnetometer boom and the antenna.

Different entry conditions: atmospheric model; entry angle; and component orientation and motion were considered. An analysis of the magnetometer boom was performed since it represents a structural type component. The results of these analyses indicate that survivability of these types of components is about the same as for Jupiter. However, the analysis on hardware containing plastic pieces indicated that these components will not receive adequate heating to ensure sterility throughout the entire material and survivability is greater than for Jupiter.

Some representative results of Saturn and Titan analyses follow. The results presented here give a sample of the types of analyses performed. Figure 1-B.1 gives some results of the analyses performed for pieces of thermal blanket. Two atmosphere models and two entry angles were considered spanning the range of extreme responses. As can be seen sterility is assured. However, in cases of tumbling at shallow angles the existence of corridors where low heating is attained was shown. The corridor exists only for pieces tumbling at a very small angle of attack with respect to the aerodynamic velocity sectors, and in cases of entry into the warm atmosphere of Saturn. Therefore, the probability of a piece of blanket layer falling within such a corridor is not high, even though it is a realistic possibility. In addition there are combinations of lift and angle of attack values that will allow entry without sterilization. All of this applies only to shallow entries into the warm atmosphere. In general, the survival of the entry by organisms residing in the blanket material can be considered small.

The analyses also showed that, in contrast to Jupiter entry, the blanket disintegration begins at a much later time due to the substantially lower entry velocity. The decomposition of the forward blanket was found to be much slower than in the case of Jupiter; and this should contribute substantially to protection of the bus from early disintegration. As noted above, only debris released early in entry would survive. Therefore, in the case of Saturn survival of organisms on debris released during breakup should be very small.

The results of the analyses for the magnetometer longeron indicate that the survivability of the longerons is unlikely. The longeron material consists of epoxy and glass fibers with epoxy completely evaporating upon attaining sterilization temperatures.

Figure 1-B.2 shows the results of the analyses for the antenna for two atmospheric models and two entry angles. The graphs show both the front and rear of the antenna giving an indication of the heat transmission across the face of the antenna. Again sterility is assured, with perhaps the exception of the portion shaded by the bus in entries into the warm atmosphere.

Large spacecraft components and pieces will result from breakup of the bus or science platform. As a sample of this a piece of bus side (aluminum) with a battery of large plastic connectors attached to the interior was selected for analysis; Fig. 1-B.3 shows a schematic of this "component." The direction of motion of the component and of the gas are shown in the figure. The heat input is at the front causing ablation of the front edge as the body travels. Figure 1-B.4 shows the results of the analyses for this component. Instead of a plot of temperature versus time, the amount of material ablated from the front by the hot gases, flowing as indicated, is shown on the vertical axis. It is seen that the effects of heating are much less in the case of Saturn than for Jupiter. The much lower entry velocity in the case of Saturn is the main contributing factor in generating these lower heat fluxes. In addition, in the Saturn warm atmosphere model the gas extends up to 996 km whereas in the case of Jupiter it extends to 635 km. This large difference provides higher ambient density at very high altitudes around Saturn causing early deceleration without generating sufficient heating to disintegrate and/or sterilize large components.

This means, in effect, that for PQ purposes the amount of encapsulated burden reduction can be considered to be close to zero. The reduction in exposed and mated burden of three orders of magnitude and two orders of magnitude, respectively, assumed for current projects appears to be reasonable.

Analyses were performed for a Titan entry, for components which did not survive Saturn entry. The spacecraft speed in the vicinity of Titan will be about 14 km/s; and because of the additional influence of Saturn, the Titan entry speed may range from 14.5 to 17.5 km/s. For the purpose of the present analysis, an entry speed of 15 km/s was assumed.

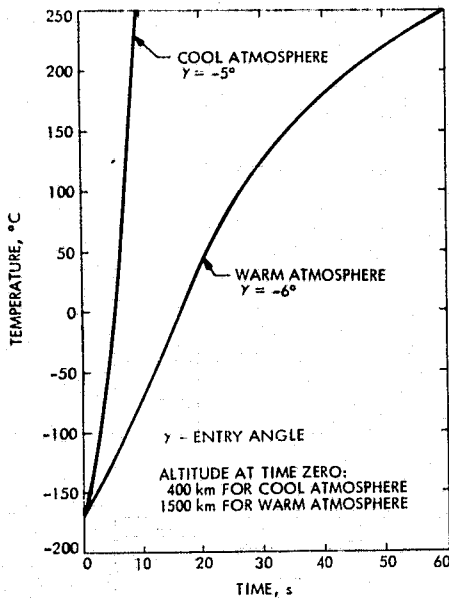


Fig. 1-B.1. Entry of aluminized mylar piece into Saturn atmosphere

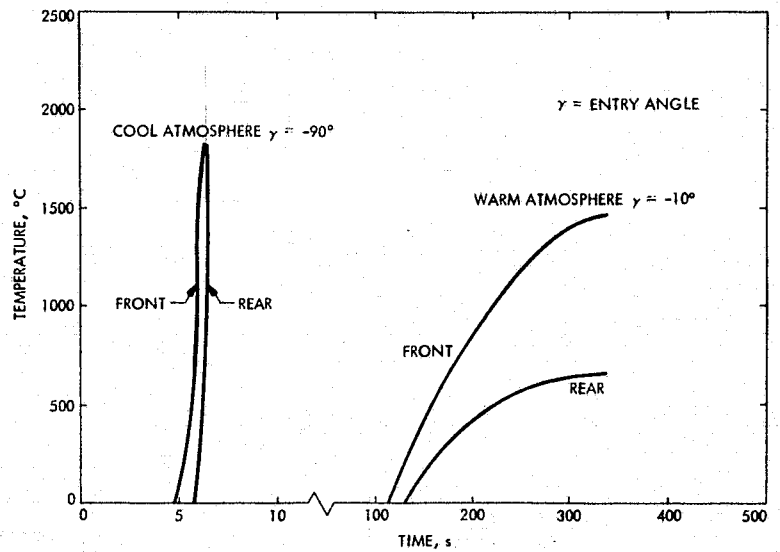


Fig. 1-B.2. Temperature history of antenna during Saturn entry

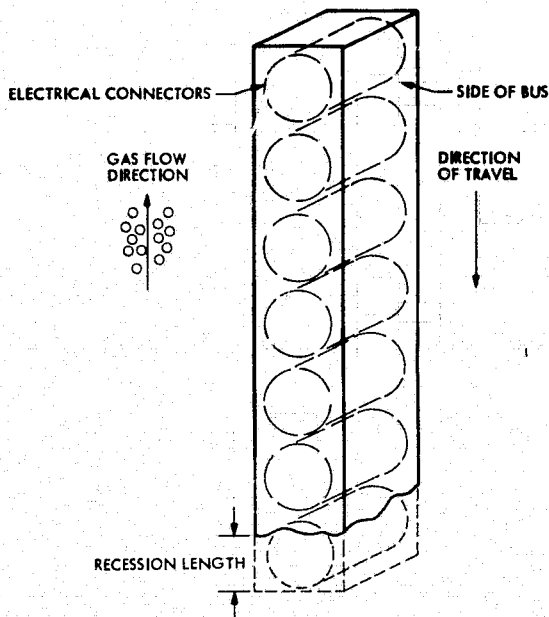


Fig. 1-B.3. Schematic of bus side and connectors

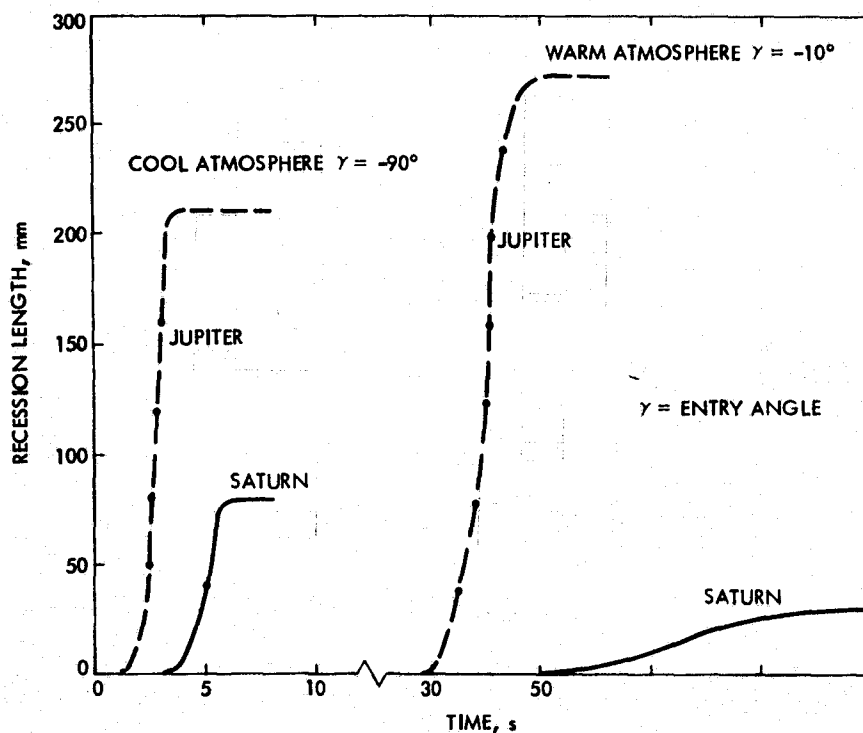


Fig. 1-B.4. Decomposition of bus side and connectors during Saturn entry

Such high entry velocity in relation to the escape velocity of only 2.7 km/s causes spacecraft skip out at high inertial entry angles; these are about -20° for the heavy atmosphere and -40° for the light atmosphere. Thus, only entries at angles higher than those stated are possible for the respective atmosphere. This situation practically precludes a step-by-step comparison of the Titan results with those of Jupiter or Saturn.

The basic premise for the analysis was that an inadvertent entry into Titan could occur when the spacecraft is aimed towards the vicinity of Saturn with a Titan flyby.

Analyses were performed for a Titan entry, for components which did not survive Saturn entry. It was found that the thermal blanket and magnetometer booms would not survive entry. In the case of the antenna only organisms on the outer portions of the antenna could be reliably assured of being sterilized. Large spacecraft components will escape substantial heating during entry.

Figure 1-B.5 shows an example of the temperature response of a small piece of mylar layer entering the light atmosphere at the inertial angle of -30° in a steady manner. When this piece is oriented at an angle of attack substantially different from flat attitude ($\mu = 90^\circ$), it skips out. It is seen that temperatures attained are high enough to achieve sterilization.

Similar results were obtained from an analysis of tumbling entries. For entries into the heavy atmosphere the calculated temperatures are much higher than those shown in the Figure 1-B.5.

Analysis to determine the thermal response of a 15-layer piece of blanket, separated from the spacecraft, indicate that it will be sterilized in all cases of entry. Should, however, thicker blankets be employed, a partial survival and inadequate heating would occur. The 15-layer thick spacecraft mounted blankets will also not survive an entry. This is due to comparatively higher ballistic coefficient at which the spacecraft will enter the atmosphere.

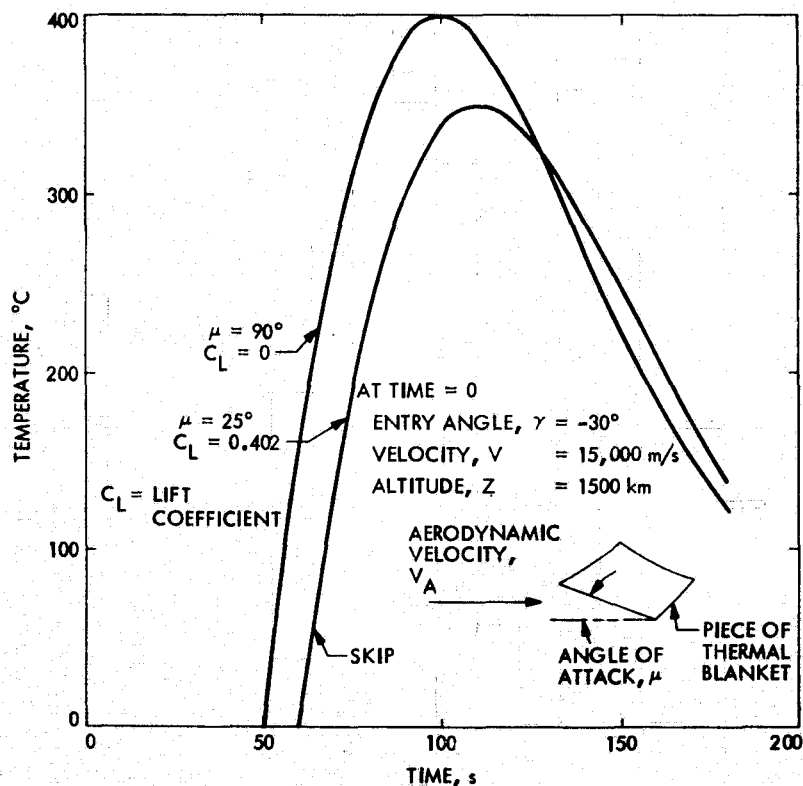


Fig. 1-B.5. Entry of aluminized mylar piece into Titan light atmosphere

Figure 1-B.6 gives the results of the magnetometer longeron analysis for two different atmospheres and entry angles and again microbial survival is unlikely.

Figure 1-B.7 shows that the outer part of the antenna will be sufficiently heated to cause sterilization. Some uncertainty does exist, however, regarding the center portion of the antenna which is shielded by the bus. Unlike the situation with Jupiter and Saturn separation of the antenna from the bus will not occur. Poor thermal conduction through the honeycomb core in the radial direction and outgassing of the face sheets may prevent adequate heat penetration towards the center of the antenna dish. Accurate analysis of this problem turns out to be very complex, and uncertainties involved do not warrant an in-depth investigation.

The results of these analyses indicate that only in the case of exposed burden can significant reduction be attributed to entry heating. Here a burden reduction of an order of magnitude can be considered reasonable.

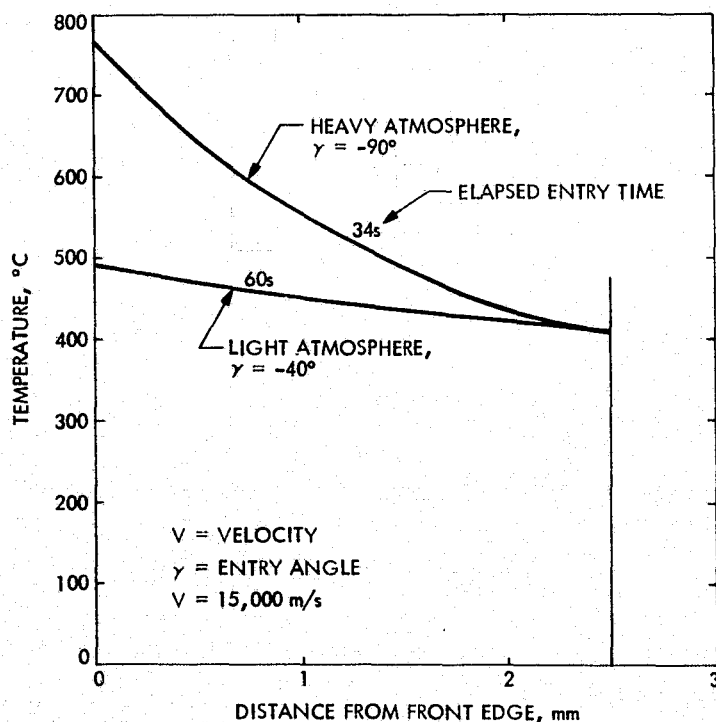


Fig. 1-B-6. Temperature profiles of magnetometer longeron during Titan entry

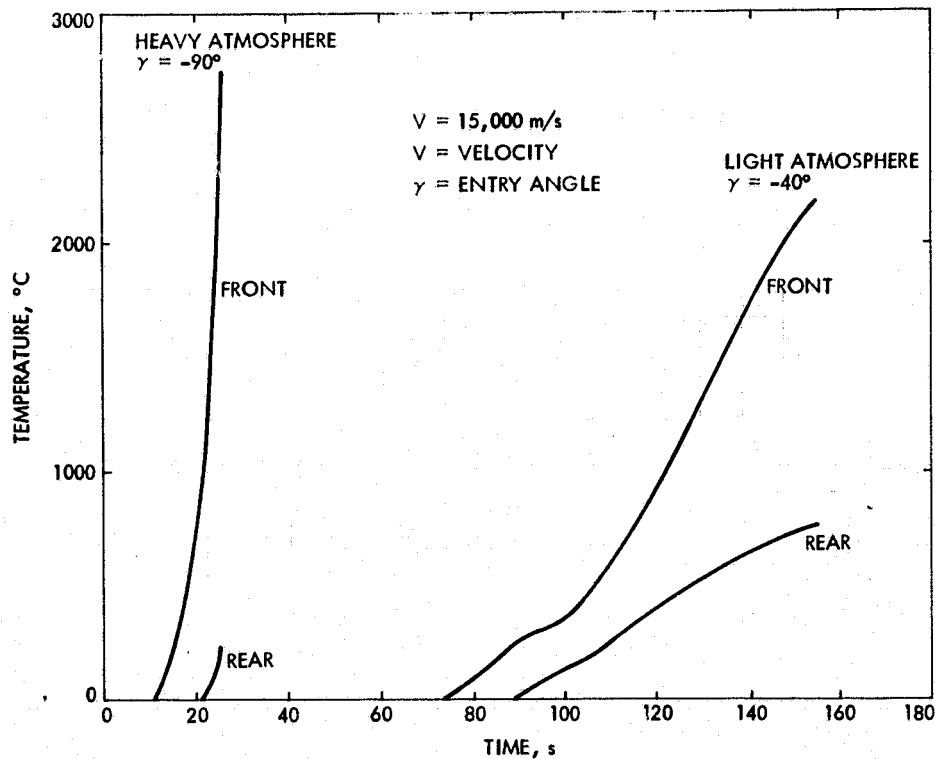


Fig. 1-B.7. Temperature history of antenna during Titan entry

1.2.2.2 Computational Tools Developed. During the course of the entry analysis task a number of analytical tools were developed, mainly in the form of computer programs.

Table 1-B.1 lists some of the major categories of programs developed and the following is a brief description of their capabilities. These tools are all in-house tools and are not currently in the form for transfer to another facility.

1. **Thermochemistry.** The purpose of these programs is to determine the state of a gas at high temperature and pressure. They were used to provide an input in the calculation of radiative flux and to provide parameters used in the various types of material ablation analyses. Among the parameters provided are the following: mole fraction of chemical species present; specific heats from frozen and equilibrium states; density; enthalpy, entropy; and molecular weight.

Table 1-B.1. Analytical Tools Developed – Computer Programs

- THERMOCHEMISTRY
- PLASMA RADIATION
- PARTICLE TRAJECTORY AND HEATING
- SPECIFIC COMPONENTS - AERODYNAMIC RESPONSE AND HEATING
 - THERMAL BLANKETS
 - ANTENNA
- MATERIAL THERMAL RESPONSE, DECOMPOSITION, AND ABLATION

2. Plasma Radiation. The program was developed for the Titan analyses and provides the radiative flux emerging from a slab of plasma.

3. Particle Trajectory and Heating. This program determines the trajectory of a particle traveling through the atmosphere and calculates the heating of these particles. The program used many input variables such as: point of release; diameter; density; drag coefficient; accommodation coefficient; emissivity; etc. It computes the particle temperature and other aspects such as skip out.

4. Specific Components – Aerodynamic Response and Heating. These programs handle the cases of specific components. They compute the motion of the component and calculate the thermal response of the component indicating temperature and ablation history. These are complex programs which handle such things as the flux of heat around an antenna dish.

5. Material Thermal Response, Decomposition and Ablation. These programs were used with components and chunks containing large amounts of plastics. They are applicable for analyzing the thermal decomposition of plastic components where the heat blockage due to ablation gases is substantial. They handle modes such as pure sublimation, melting, charring, and surface or in-depth decomposition. Convective as well as radiative heat blockages are fully and properly accounted for.

1.2.3 Future Activities

No future activity is planned.

1.2.4 Presentations

Gonzalez, C., "Jupiter Entry Analysis," presented at NASA Spacecraft Sterilization Technology Seminar, Cocoa Beach, Florida, August, 1975.

1.2.5 References

1. Planetary Quarantine Annual Review, Space Research and Technology, July 71 - July 72, 900-597, February 1973
2. Planetary Quarantine Semi-Annual Review, Space Research and Technology, 1 July - 31 December 1972, 900-608.
3. Planetary Quarantine Semi-Annual Review, Space Research and Technology, 1 July - 31 December 1973, 900-655, April, 1974
4. Planetary Quarantine Semi-Annual Review, Space Research and Technology, 1 January - 30 June 1974, 900-675, September 30, 1974
5. Planetary Quarantine Semi-Annual Review, Space Research and Technology, 1 July - 31 December 1974, 900-701, April 18, 1975

SECTION II

NATURAL SPACE ENVIRONMENT STUDIES
(NASA NO. 193-58-61-02)ContentsTitle and Related PersonnelSubtask A
para. 2.1EFFECT OF PLANETARY TRAPPED
RADIATION BELT ON MICROORGANISMSCognizance: D. Taylor
J. BarengoltzAssociate
Personnel: C. Hagen (Bionetics)Subtask B
para. 2.2EFFECT OF SOLAR WIND ON
MICROORGANISMS

Cognizance: J. Barengoltz

Associate
Personnel: J. Farber (Bionetics)
S. Donovan (Bionetics)
A. Ferreira (Bionetics)
S. Yamada (Bionetics)
C. Hagen (Bionetics)Subtask C
para. 2.3EFFECT OF SPACE VACUUM ON
MICROORGANISMSCognizance: M. Wardle
C. Hagen (Bionetics)Associate
Personnel: E. Hagen
A. Ferreira (Bionetics)Subtask D
para. 2.4PROBABILITY OF GROWTH IN PLANETARY
ATMOSPHERE AND SATELLITESCognizance: D. Taylor
A. IngersollAssociate
Personnel: C. Myers
N. Divine

2.1 EFFECT OF PLANETARY TRAPPED RADIATION BELT ON MICROORGANISMS

2.1.1 Subtask A Introduction

The objective of this subtask is to determine the effect of planetary trapped radiation belts on the survival of microorganisms associated with an unsterile spacecraft.

With flyby missions now planned for Jupiter and Saturn and possible Jupiter orbiters and probes, the trapped radiation belts may represent an environment lethal to microorganisms and thereby reduce the requirements for decontamination of spacecraft before launch.

The major components of planetary trapped radiation belts are electrons and protons. The approach of the present task is to evaluate possible biological effects of these belts by subjecting spacecraft microbial isolates to different energies, exposures, and dose rates of those particles.

2.1.2 Significant Accomplishments

A procurement for a study to determine the effect of particle radiation on encapsulated microorganisms has been initiated. This study will provide data to be compared with data for microbes on a surface, obtained previously under this task. The effect of factors not modelled in conventional radiation transport calculations will be investigated.

An electron Monte Carlo transport computer program has been modified and adapted for use on the JPL Univac 1108 computer system.

2.1.3 Future Activities

The contracted study on encapsulated microorganisms will be performed. The data obtained will be analyzed and compared with results for microbes on a surface.

The present radiation environment for Jupiter orbits is provisional and will be recomputed as Pioneer 11 radiation measurements are incorporated into the radiation belt models. These updated models will then allow an evaluation of external radiation environments in terms of electron test data.

An analysis of the effect of secondary radiation will be performed with the use of the new models, the encapsulated microbe data, and the electron transport program.

Planning for the procurement of a study with high energy protons similar to those present in planetary trapped radiation belts will continue.

2.2 EFFECT OF SOLAR WIND RADIATION ON MICROORGANISMS

2.2.1 Subtask B Introduction

The objective of this subtask is to determine the effect of solar wind radiation on microorganisms associated with nonsterile spacecraft.

This study is directed towards determining the reduction in spacecraft-associated microbial burden attributable to solar wind radiation. The data obtained will be utilized to update probability constants in the assessment of mission planetary quarantine constraints.

2.2.2 Approach

In order to fulfill the objectives of this task, an initial test program to investigate the effect of solar wind electrons on test microorganisms held in a vacuum has been established. A literature survey indicates biological effectiveness for electrons with energy in excess of 1 keV. Measurements and models of the solar wind electron spectrum imply an upper limit to the energy range of interest at about 5 keV. Parametric tests in this energy range, 1-5 keV, will be conducted at accelerated dose rates to permit typical mission doses (fluences) in acceptable test durations. Other radiation effects data indicate significant dose rate effects are unlikely. At each energy tests with varying doses will be performed to obtain survival curves.

The first formal experimental phase will consist of tests with MM'71 isolates (sporeformers and non-sporeformers) and Staphylococcus epidermidis and spores of Bacillus subtilis var. niger as comparative organisms. A second phase will be an analogous program with naturally occurring microbial populations as samples.

In long range planning with the electron source, close-to-real time exposures and sequential exposures with varying energy to simulate the spectrum are being considered.

2.2.3 Significant Accomplishments

2.2.3.1 The Solar Wind Electron Source (SWES). The SWES system, consisting of an electron gun subsystem, a dosimetry subsystem, and a vacuum subsystem, has been described in detail in a previous JPL Semi-Annual Review, April, 1975 (Ref. 1).

2.2.3.2 Test Conditions for Cultured Organisms. The irradiations of the pure cultured organisms have been conducted in vacuum ($<10^{-5}$ torr) at room temperature according to the test matrix Table 2-B.1. The electron fluences (doses) have been selected to include predicted fluences for a 1 year period at 1 AU heliocentric distance in an electron energy histogram, Table 2-B.2. Note that the test fluences decrease sharply with increasing energy to reflect the actual power law flux. The test fluxes (dose rates) represent a large acceleration over the models for the environment.

2.2.3.3 Interim Results for Cultured Organisms. At this writing the formal test program for cultured organisms, consisting of four replicate exposures of each matrix entry, Table 2-B.1, is nearly complete. The electron dosimetry procedures and microbiological preparation and assay procedures have been reported in Reference 1.

The data reduction has been automated as follows: The raw data from each experimental run is written on coding forms from which computer cards are keypunched. An initial program reduces this data in the form of a printed run result summary and an output card deck containing the logarithm of the survival fraction and standard deviation for each organism tested. A statistical quality factor for the entire run is also calculated. A collection of such output decks, representing a specified number of replicate runs and fluence values, is sorted and ordered by a second computer program. This program calculates the mean survival fraction (over replicate runs) and standard deviation for each organism as a function of fluence. The mean spore and mean non-sporeformer isolate survival fractions are also calculated. These results are then printed in a matrix for each particle type, flux, and energy in a determined order. A sample output page is shown in Fig. 2-B.1. The program finally performs a

Table 2-B.1. Electron Test Matrix for Cultured Organisms

| Energy | Flux (e/cm ² s) | | | | | |
|---------|-------------------------------|---------------------------------|----------------------|----------------------|----------------------|----------------------|
| 1.5 keV | 10 ¹² | Fluence (e/cm ²) | 5 x 10 ¹³ | 1 x 10 ¹⁴ | 5 x 10 ¹⁴ | 1 x 10 ¹⁵ |
| | | Duration | 50s | 100s | 500s | 1000s |
| 2.0 keV | 5 x 10 ¹⁰ | Fluence (e/cm ²) | 2 x 10 ¹² | 1 x 10 ¹³ | 5 x 10 ¹³ | 2 x 10 ¹⁴ |
| | | Duration | 40s | 200s | 1000s | 4000s |
| 3.0 keV | 10 ¹⁰ | Fluence (e/cm ²) | 5 x 10 ¹¹ | 2 x 10 ¹² | 5 x 10 ¹² | 1 x 10 ¹³ |
| | | Duration | 50s | 200s | 500s | 1000s |
| 4.5 keV | 5 x 10 ⁹ | Fluence (e/cm ²) | 2 x 10 ¹¹ | 5 x 10 ¹¹ | 2 x 10 ¹² | 5 x 10 ¹² |
| | | Duration | 40s | 100s | 400s | 1000s |

Table 2-B.2. Predicted Solar Wind Electron Fluence for 1 Year at 1 AU

| Energy Interval keV | Model A Fluence ^a cm ⁻² | Model B Fluence ^b cm ⁻² |
|---|--|--|
| 1.0 - 2.5 | 3 x 10 ¹² | 6 x 10 ¹² |
| 2.5 - 4.0 | 4 x 10 ¹¹ | 9 x 10 ¹¹ |
| 4.0 - 5.0 | 9 x 10 ¹⁰ | 2 x 10 ¹¹ |
| ^a based on: Divine, T. N., "Interplanetary Charged Particle Environments," JPL TM 33-637 (1973). ^b based on: Montgomery, M. D., Bame, S. J., and Hundhausen, A. J., "Solar Wind Electrons: Vela 4 Measurements," Journal of Geophysical Research, Space Physics <u>73</u> , No. 15, pp. 4999-5003 (1968). | | |

1.5 KEV ELECTRON
AT A FLUX OF $1.0 \times 10^{12} (\text{CM}^{-2} \text{ SEC})^{-1}$

TABLE OF SURVIVAL FRACTIONS (WITH S.D.)

ORGANISM / FLUENCE (CM^{-2})

| | 5.00×10^{13} | | 1.00×10^{14} | | 5.00×10^{14} | | 1.00×10^{15} | |
|------------|-----------------------|----------|-----------------------|----------|-----------------------|-----------|-----------------------|-----------|
| 4 | .63+00 | (.18+00) | .96-01 | (.20-01) | .32-01 | (.16-01) | .38-01 | (.31-01) |
| 5 | .58+00 | (.14-01) | .31+00 | (.14-01) | .12+00 | (.37-01) | .78-01 | (.45-01) |
| SE | .10+01 | (.42+00) | .79-01 | (.72-01) | .00 | (-.10+01) | .00 | (-.10+01) |
| 1 | .85+00 | (.18+00) | .36+00 | (.71-01) | .10+00 | (.52-01) | .71-01 | (.25-01) |
| 2 | .11+01 | (.31+00) | .48+00 | (.85-01) | .49-01 | (.71-03) | .64-01 | (.50-01) |
| 9 | .47+00 | (.19+00) | .14+00 | (.71-02) | .31-01 | (.21+02) | .10-01 | (.25-02) |
| 13 | .41+00 | (.18+00) | .11+00 | (.14-01) | .57-01 | (.11-01) | .20-01 | (.78-02) |
| 16 | .71+00 | (.71-01) | .31+00 | (.22+00) | .56-01 | (.39-01) | .25-01 | (.16-01) |
| BSN | .38+00 | (.49-01) | .85-01 | (.21-01) | .72-02 | (.57-03) | .38-02 | (.14-03) |
| SPORE MEAN | .70+00 | (.49+00) | .28+00 | (.21+00) | .70-01 | (.61-01) | .38-01 | (.44-01) |
| VEG MEAN | .61+00 | (.17+00) | .20+00 | (.43-01) | .78-01 | (.47-01) | .58-01 | (.63-01) |

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900-715

Fig. 2-B. 1. Sample data summary (computer output) for 1.5 keV based on two replicate runs.
Negative standard deviations indicate data not available

linear regression on the logarithm of the survival fraction versus fluence for each organism and optionally produces plots of the mean reduced data.

Interim results based on two replicates are shown for all energies tested in Figs. 2-B.2, 3, 4, 5, 6, 7, 8, and 9. Although these results will be replaced by better statistics when the formal program is completed, certain features may be noted: (1) There is a large variation in the response for different organisms. This variation is reflected in the mean spore and mean non-sporeformer survival fractions. (2) Isolate #5, a micrococcus, is extremely resistant compared to isolate #4, a micrococcus, or Staphylococcus epidermidis. This factor causes rather large mean non-sporeformer survival fractions (Figs. 3, 5, 7, and 9). (3) An obvious plateau effect at high fluences occurs for survival fractions between 0.001 and 0.01 for spores and about 0.1 for the mean non-sporeformers.

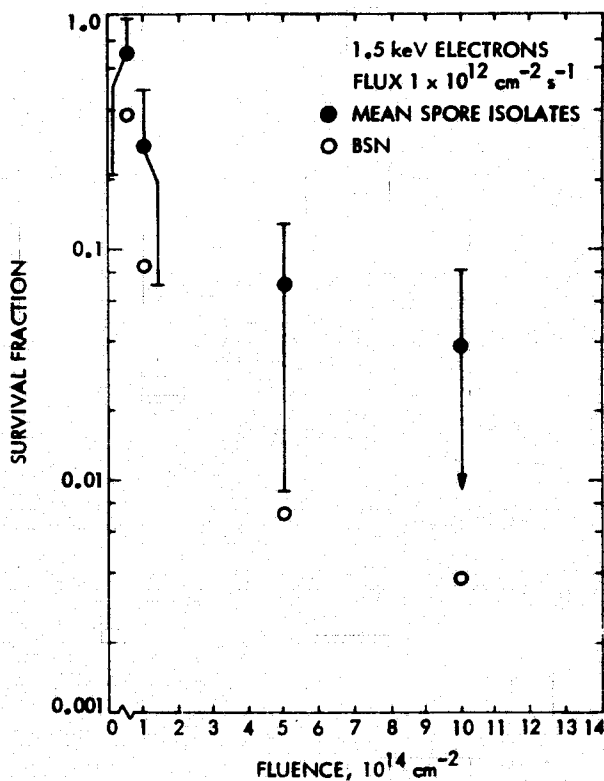


Fig. 2-B.2. Spore survival at 1.5 keV electron energy (based on two replicate runs)

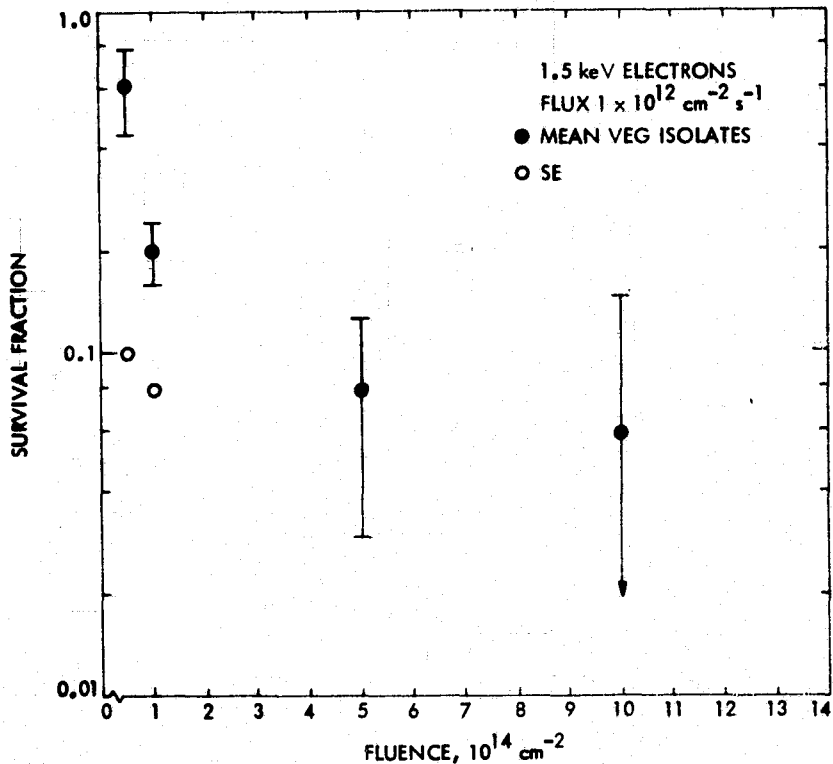


Fig. 2-B.3. Nonsporeformer survival at 1.5 keV electron energy (based on two replicate runs)

The plateau effect is tentatively explained by sample clumping and the resultant shielding of individual organisms. It is well known that electrons in the energy range studied cannot penetrate through an organism, e.g., the range of a 5 keV electron in organic material is only $0.57 \mu\text{m}$ (Ref. 2 and 3). Under this hypothesis, the minimum survival fraction is then interpreted as the fraction of cells which are shielded by at least one other cell. Since the test samples represent an extremely large density ($\sim 10^6 \text{ cm}^{-2}$) compared to a real surface, this effect would render the present results very conservative.

Isolate #5, a micrococcus in a tetrad form, may be an exception in that any cell of the tetrad will typically be shielded by one or more of the others. This self-shielding may explain both the resistance of #5 to the radiation and the high plateau survival fraction for the mean non-sporeformers, dominated by the results for #5.

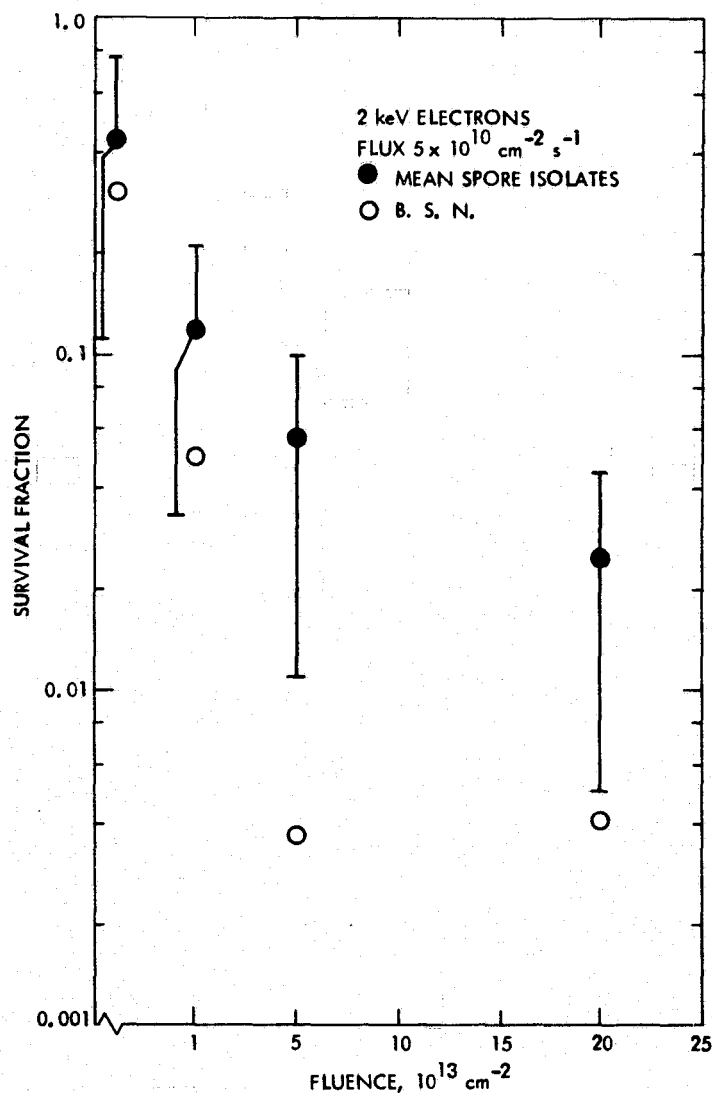


Fig. 2-B.4. Spore survival at 2.0 keV electron energy (based on two replicate runs)

The results may also be summarized in terms of the D_{10} values, or the fluence required to produce a one order of magnitude reduction in the survival fraction. Calculated values from the linear regression analysis for four fluence values are given in Fig. 2-B.10. Because of the plateauing effect the D_{10} values represent over-estimates of the fluence required for the first order of magnitude reduction. Note that D_{10} for *B. subtilis* as measured by Davis (Ref. 4) is considerably smaller than the present results, especially at the

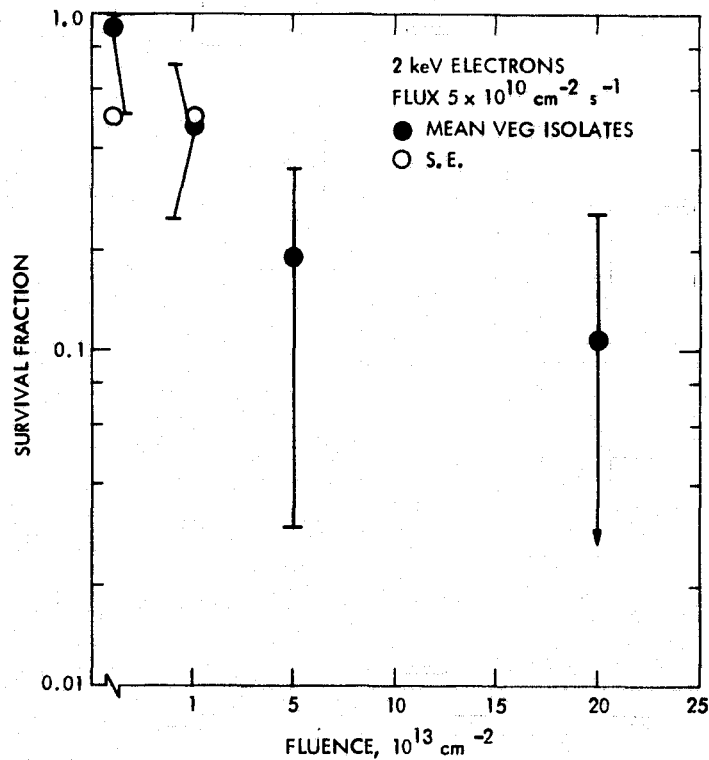


Fig. 2-B.5. Nonsporeformer survival at 2.0 keV electron energy (based on two replicate runs)

higher energies. Davis' values are for the first order of magnitude reduction since she inoculated only 50 microbes on a 1 cm^2 substrate to avoid clumping. A revision of our D_{10} values, to a fit for survival fractions greater than 0.02 only, yields excellent agreement with the published work.

2.2.4 Future Activities

The formal tests with pure cultured organisms will be completed. The data will be reduced and summarized.

Formal tests with naturally occurring organisms collected on fall-out plates will be conducted.

A study of the shielding factor discussed above will be undertaken. Plans call for more dilute inoculations at the higher fluences and exposures at higher (more penetrating) electron energies.

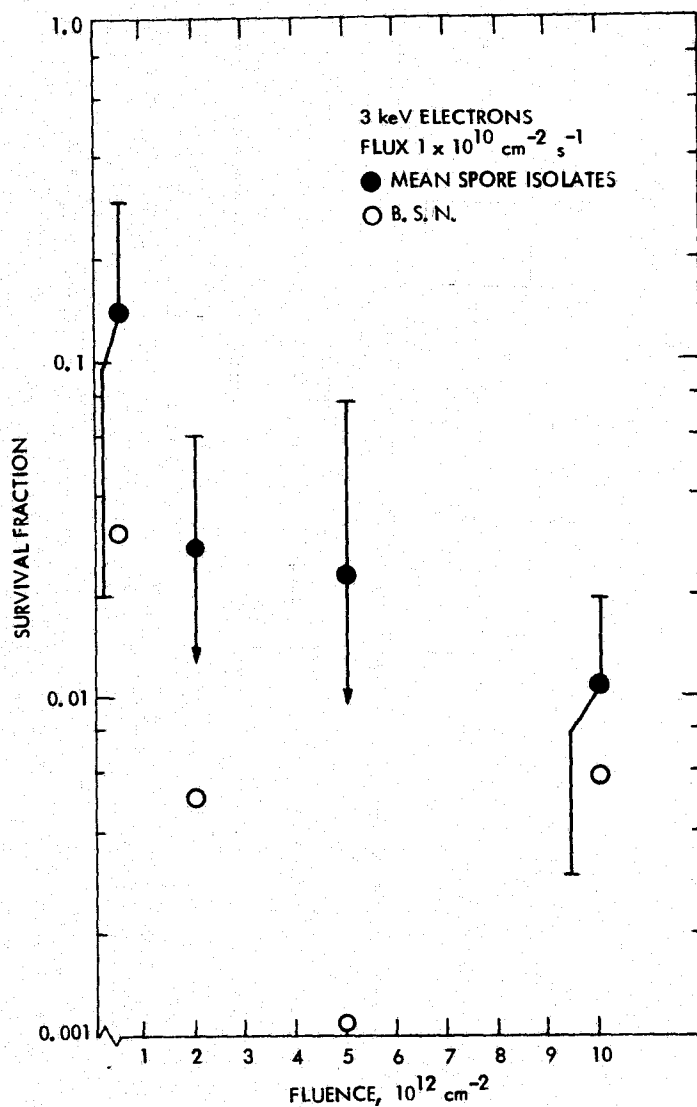


Fig. 2-B.6. Spore survival at 3.0 keV electron energy (based on two replicate runs)

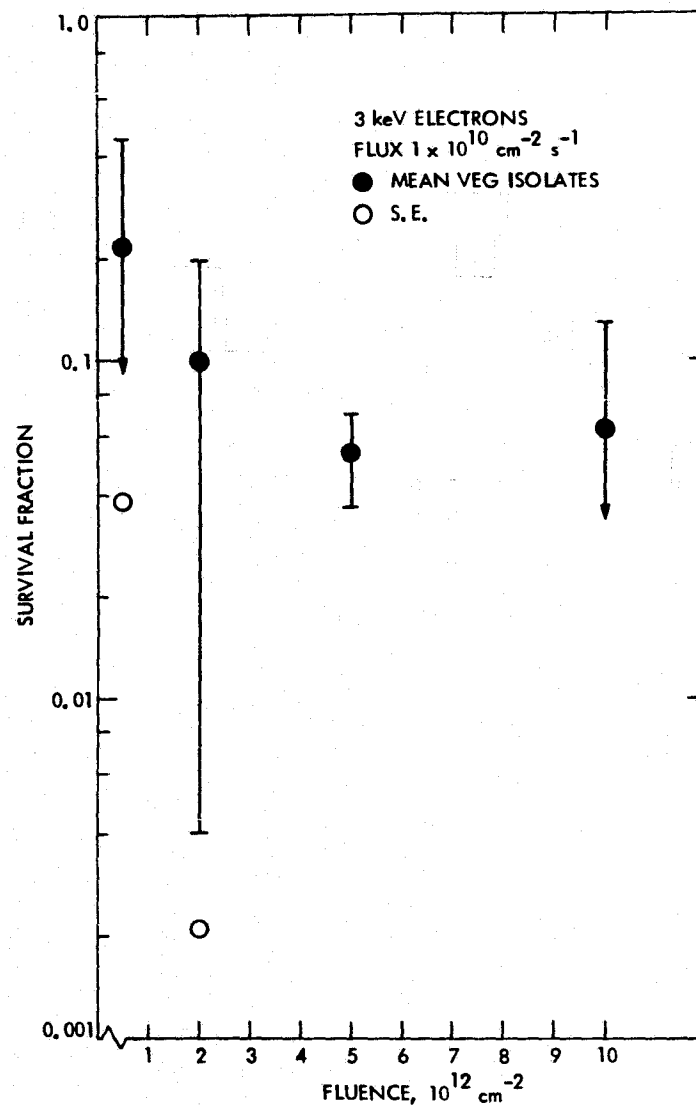


Fig. 2-B.7. Nonsporeformer survival at 3.0 keV electron energy (based on two replicate runs)

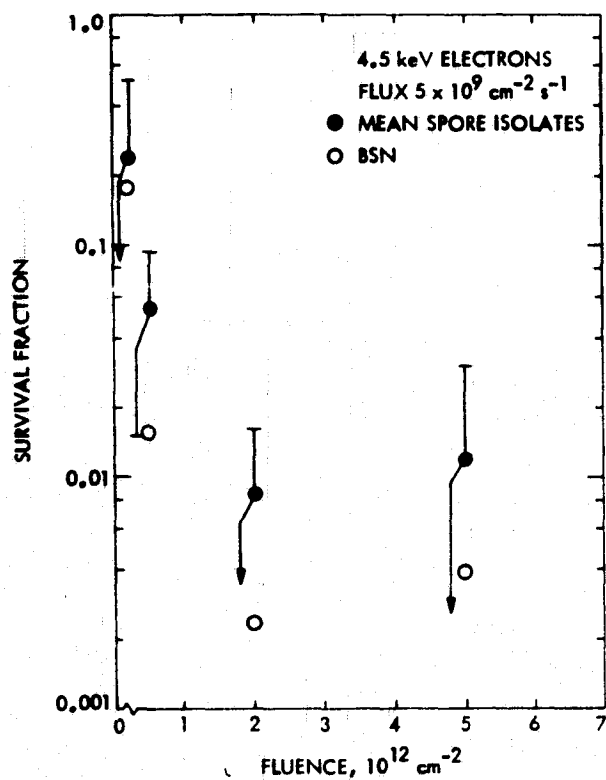


Fig. 2-B.8. Spore survival at 4.5 keV electron energy (based on two replicate runs)

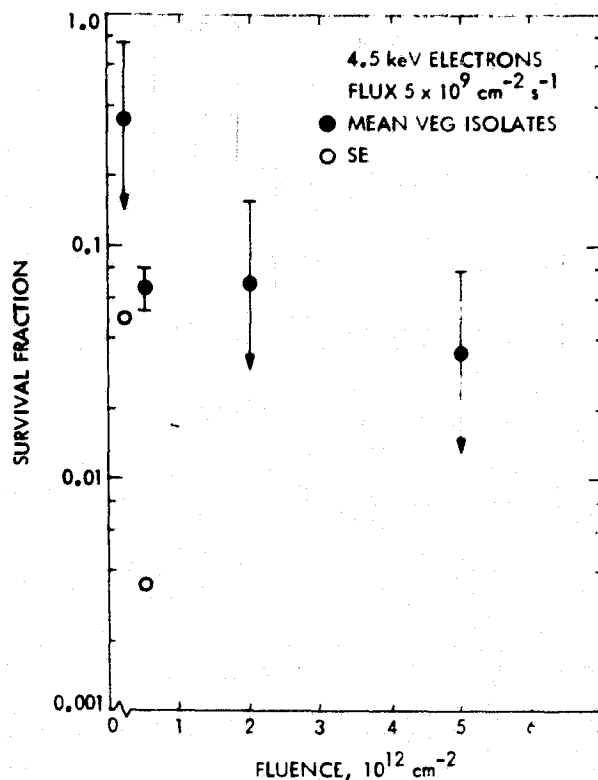


Fig. 2-B.9. Nonsporeformer survival at 4.5 keV electron energy (based on two replicate runs)

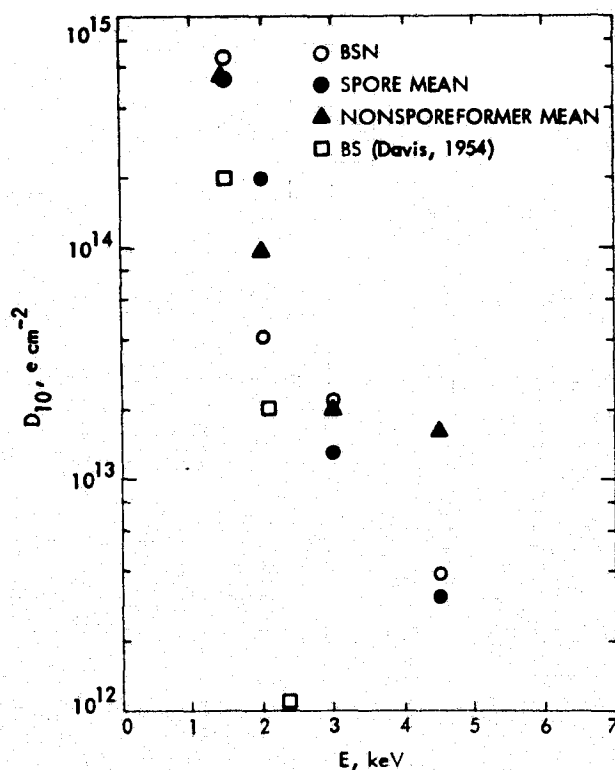


Fig. 2-B.10. D_{10} values as a function of electron energy (Note: Davis' data based on inoculation of 50 microbes/ cm^2)

2.2.5 Presentations

Barengoltz, J., "Effect of Solar Wind Radiation on Microorganisms," presented at NASA Spacecraft Technology Seminar, Cocoa Beach, Florida, August 1975.

2.2.6 References

1. Planetary Quarantine Semi-Annual Review, Supporting Research and Technology, 1 July - 31 December 1974, JPL Doc. 900-701, pp. 2-3 to 14 (1975).
2. Lea, D. E., The Action of Radiation on Living Cells, Cambridge University Press (1947).
3. Davis, M., "Range Measurements of Low Voltage Electrons," Physical Review 94, p. 243 (1954).
4. Davis, M., Arch. Biochem. and Biophys. 48, pp. 469-481 (1954).

2.3 EFFECT OF SPACE VACUUM ON MICROORGANISMS

2.3.1 Subtask C Introduction

The objective of this task is to determine the effects of extended exposure to space vacuum and spacecraft temperatures on the survival of microbial species representative of those found on spacecraft.

2.3.2 Significant Accomplishments

Work has commenced to determine the thermal vacuum resistance of naturally occurring microbial populations. Efforts to date have primarily involved the engineering checkout of the test system. Initial microbiological tests are underway.

2.3.3 Future Activities

Naturally occurring microbial populations will be tested for survival in a vacuum of approximately 2×10^{-6} torr at temperatures of 55, 65 and 75°C for periods up to 28 days.

2.4 PROBABILITY OF GROWTH IN PLANETARY ATMOSPHERES AND SATELLITES

2.4.1 Subtask D Introduction

The objectives of this subtask are to relate environmental parameters affecting microbial growth to conditions present in the atmospheres of Jupiter and Saturn, and to identify and study satellites of Jupiter and Saturn having possible biological interest.

2.4.2 Significant Accomplishments

2.4.2.1 Dynamics of Jupiter's Atmosphere. The material reported here extends earlier work reported for this subtask, namely the existence of layers in Jupiter's atmosphere within which pressure, temperature, and water activity are suitable for the survival and growth of terrestrial microorganisms. It addresses the likely residence times of anaerobic organisms within the hospitable layers.

A contract is now in progress with Dr. Andrew P. Ingersoll (Division of Geological and Planetary Sciences, California Institute of Technology) whose overall objective is the development of a working numerical model of Jupiter's atmosphere to pressures up to 100 bars. The model will be suitable for use in determining orbital lifetimes, entry probe deceleration and heating, and transport, survival and growth of anaerobic organisms.

Under this contract a thorough review paper (Ingersoll, 1975) has been completed and submitted for publication in Space Science Reviews. It includes current information on the neutral atmosphere of Jupiter, with approximately equal emphasis on composition and thermal structure on the one hand, and markings and dynamics on the other. Studies based on Pioneer 10 and 11 infrared data are used to refine the atmospheric model. Data on the interior are reviewed for the information they provide on the deep atmosphere. The markings and dynamics are discussed with emphasis on qualitative relationships and analogies with phenomena in the Earth's atmosphere.

The vertical temperature structure is remarkably uniform over the planet, such that the effective temperature and emergent flux (internal and re-radiated solar power) are constant with latitude and longitude to within a few percent. A typical temperature profile, and the wavelength ranges from which the segments are inferred, is shown in Fig. 2-D. 1, and beside it is diagrammed a theoretical, three-layer cloud structure. However, there are small horizontal variations in the vertical temperature profile which are long-lived because of their relationship to local cloud formation and dissipation processes, reflection and absorption of solar radiation, and properties observed and inferred for the dynamics. The observed bands paralleled to Jupiter's equator are manifestations of the above items, and exhibit obvious differences in visual albedo and in infrared brightness temperatures (at many, but not all, wavelengths). In particular, the uppermost (NH_3) cloud deck is commonly present in bright bands named zones (and in the Great Red Spot) and absent in darker bands named belts. A schematic of the temperatures, circulation and cloud processes in these bands is shown in Fig. 2-D. 2.

As suggested above, the dynamics of these two kinds of bands are very distinct, and preliminary calculations have been completed for some pertinent properties. The zones and red spots are mixed areas in which active convection occurs, and mixing-length theory (developed for the analysis of convection in stellar interiors and envelopes) has been applied, leading to residence times of the order of a day in the inter-cloud regions (e. g., altitudes 60 to 120 km in Fig. 2-D. 1). The belts are unmixed areas in which the absence of turbulent convection allows cloud dissipation, and meteorological calculations (developed for the Earth's Trade-Wind belts) have been applied, leading to residence times greater than or of the order of 0.1 year.

2.4.3 Future Activities

Several pertinent dynamical mechanism will be used to parametrize the small- and intermediate-scale motions in the numerical model of the largest-scale motions. Documentation of the details and conclusions of the

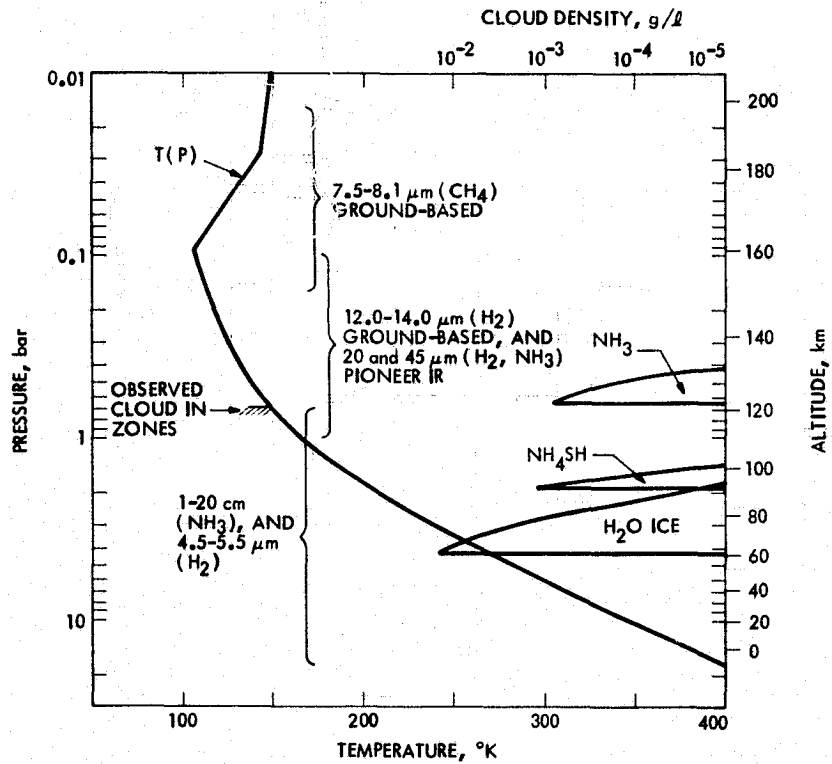


Fig. 2-D.1. Jupiter atmosphere temperature and cloud profiles

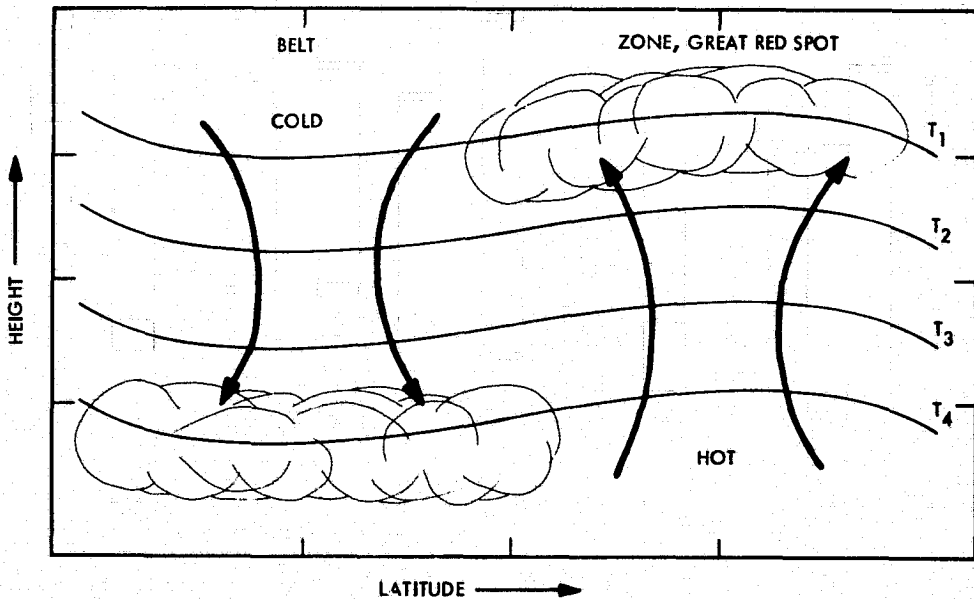


Fig. 2-D.2. Schematic of belt-zone circulation

atmospheric profiles, residence time considerations, and numerical modeling results will be completed by the end of the contract (31 December 1975).

2.4.4 Presentations

Ingersoll, A., "Jupiter Dynamic Atmosphere Study, " presented at NASA Spacecraft Sterilization Technology Seminar, Cocoa Beach, Florida, 12-13 August 1975.

2.4.5 Publications

Ingersoll, Andrew P., 1975: "The Atmosphere of Jupiter, " accepted for publication in Space Science Reviews.

2.5 EFFECT OF SOLAR ELECTROMAGNETIC RADIATION ON MICROORGANISMS

2.5.1 Subtask E Introduction

The objective of this task is to estimate the effect of solar electromagnetic radiation (SER) on the survival of microbial populations in a space environment. Efforts will be addressed to the investigation of the photobiological effect of SER in a fashion that permits direct transference of the results to considerations of planetary quarantine. Such information will enable the updating of probability constants in the assessment of applicable planetary quarantine constraints for a mission.

2.5.2 Approach

The approach for this task involves the subjection of test species to SER in a manner that will yield interpretive data on the response of spacecraft biocontaminants to the SER of space. Primarily, this entails the high vacuum irradiation of microorganisms, pure cultured and naturally occurring, with broad spectrum SER (far ultraviolet through infrared).

Pure cultured species are studied to define the effect of SER under different dose, dose rate and temperature conditions. Seven isolates (5 spore-formers and 2 nonsporeformers) from Mariner Mars 1971, Staphylococcus epidermidis and spores of Bacillus subtilis var. niger (BSN) are tested. Irradiation is conducted at solar constants* (dose rates) of 0.1, 0.5 and 1.0 sun with temperatures at irradiation of -125, -15 and 70°C, respectively. These conditions respectively correspond to representative near Jupiter, Mars and Earth environments. In order to obtain survivor curves, organisms are exposed to varying doses for each dose rate - temperature condition employed.

Naturally occurring populations are collected on Viking type solar cell fixtures and exposed to SER in a natural state; i. e., no laboratory treatment of the organisms is instituted prior to test environment exposure. As with

*For this study, 1.0 solar constant is defined as a beam intensity, at the plane of irradiance, of 0.54 mW cm^{-2} in the wavelength interval from 200 to 270 nanometers.

axenic cultures, dose, dose rate and temperature are imposed as experimental variables.

2.5.3 Significant Accomplishments

2.5.3.1 Solar Electromagnetic Radiation Test System. A complete account of the solar electromagnetic test system is given in the JPL-PQ Semi Annual, Sept. 30, 1974.

2.5.3.2 Experimental Activities.

1. Pure cultured microorganisms. A complete account of the preparation, irradiation and assay of pure cultured organisms is given in the JPL-PQ Semi Annual, Sept. 30, 1974.

2. Naturally Occurring Microorganisms. A complete account of the collection, irradiation and assay of naturally occurring microbial populations is presented in the JPL-PQ Semi Annual, April 30, 1975.

3. Test Results. Tests were completed on the effect of SER on pure cultured microorganisms under 0.5 sun, -15°C conditions. The spore results shown in Fig. 2-E.1 represent composite means of 5 MM'71 isolates and BSN; two of the nonsporeformers (Micrococcus (Type II) and S. epidermidis) were totally inactivated at very low doses (200 ergs mm^{-2} or less) therefore results are plotted for only one nonsporeformer, Micrococcus (Type VI). The major inactivation effect on both classes of organism was seen to occur within the first $2,000 \text{ ergs mm}^{-2}$ of irradiation. As in previously reported tests conducted at 0.1 and 1.0 sun, the spores showed significantly greater survival than did nonsporeformers at doses in the $4,000$ to $8,000 \text{ ergs mm}^{-2}$ range.

Figure 2-E.2 shows the results to date on the effect of SER (0.5 sun, 65°C) on naturally occurring microbial populations. These data represent survival fractions for populations collected on solar panels exposed for 5 to 7 days to a spacecraft component test facility. As discussed in the previous Semi Annual, (April 30, 1975) certain similarities of these data with those

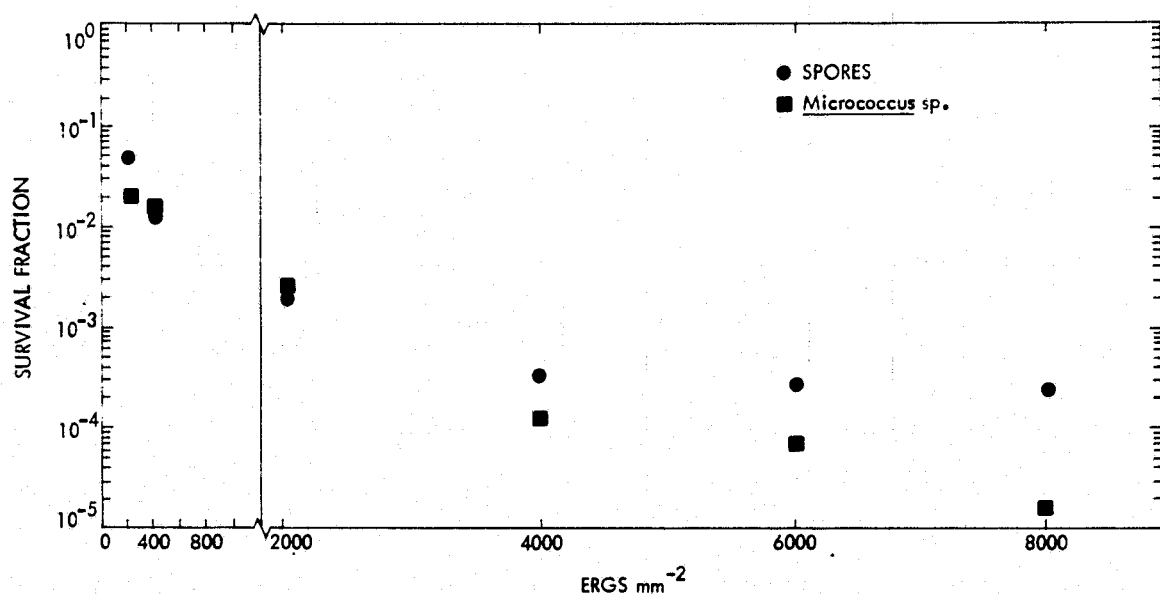


Fig. 2-E. 1. Effect of SER at 0.5 Sun, -15°C on pure cultured populations

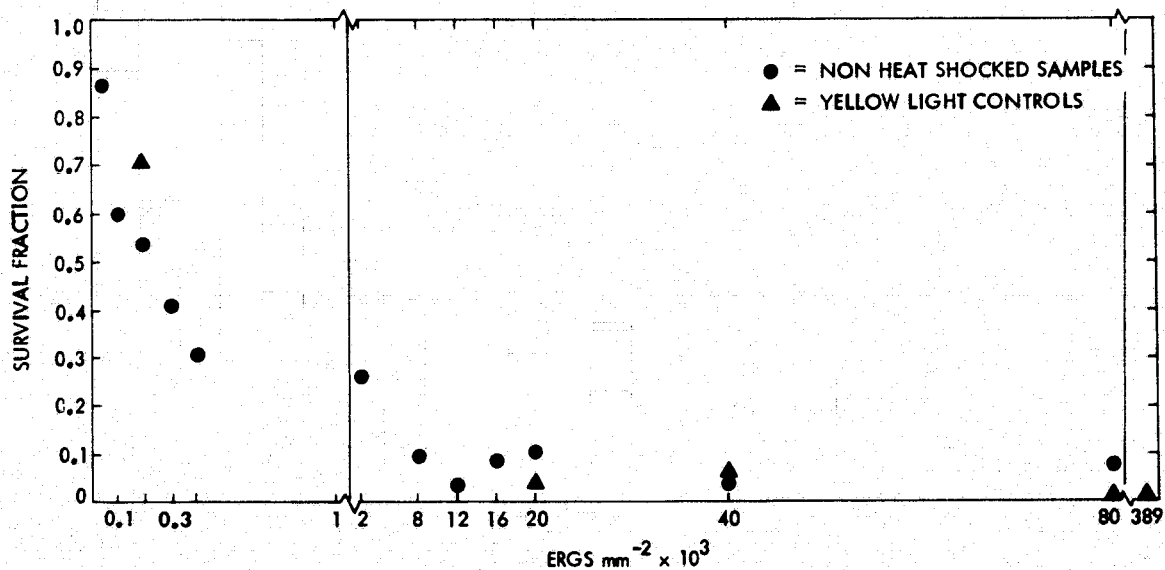


Fig. 2-E.2. Effect of SER at 0.5 Sun, 65°C on naturally occurring populations

acquired from pure cultured studies exist; i. e., the major inactivation within the first 2,000 ergs mm^{-2} and the plateauing of the survivor curve, thereafter. On the other hand, the greater resistance of the naturally occurring populations prompted an examination of photoreactivation (PHR) as an experimental variable that could have contributed to an increased survivorship for naturally occurring populations.

It is conceivable that samples exposed to SER could experience PHR upon subsequent exposure to laboratory fluorescent lighting. The literature seems to indicate this to have been unlikely in our pure culture tests and our own examination of PHR of pure cultured organisms as a consequence of our assay protocol corroborated this view. However, no applicable literature on PHR of naturally occurring organisms is available. Therefore, tests were conducted to evaluate PHR as a phenomenon leading to the higher observed survival fractions for naturally occurring microbes. This was accomplished by processing all samples in a yellow light environment subsequent to SER exposure. (Yellow light is composed of wavelengths of approximately 560-590 nm; the most significant wavelength range for PHR is 300-500 nm). The triangles in Fig. 2-E.2 represent survival fractions obtained when tests were conducted in this manner. If PHR were a significant enhancer of the survival fractions one would expect the yellow light tests to show smaller survival fractions; no trend in this regard was noted thus leading to the conclusion that PHR was not a significant variable in our experimental protocol. Therefore, the greater resistance of naturally occurring microbes is most likely due to non-viable particulate shielding and/or a greater innate resistance of these populations to SER. (When a yellow light test was run with a dose of 3.89×10^5 ergs mm^{-2} a survival fraction of 0.014 was observed (Fig. 2-E-2); survival fractions for pure cultured organisms were approximately 1 to 3 orders of magnitude less than this for doses of 8×10^3 ergs mm^{-2} .)

Control population levels averaged approximately 1.4×10^4 colony forming units ft^{-2} for non heat shocked and 2.6×10^2 colony forming units ft^{-2} for heat shocked (post-irradiation exposure to 80°C for 18 minutes in a water bath) naturally occurring population samples. The ratio of heat shocked to non heat shocked counts for control samples was approximately one-half that observed for irradiated samples (0.011 vs 0.020). However, despite the

apparent greater relative resistance of the heat shocked organisms to SER, no consistent trend in the data was noted; i. e., what appeared to be a sterilizing dose under particular test conditions often yielded a significant number of survivors when the test was repeated. This inconsistency was probably a function of the low population numbers and a correspondingly greater impact of random particle shielding on survival fractions.

Survivors of naturally occurring tests in the plateau range (2.0×10^4 to 3.9×10^5 ergs mm^{-2}) were identified. Of 35 randomly selected isolates, 26 were categorized as nonsporeformers and 9 as sporeformers. Thus, naturally occurring nonsporeformers were observed to survive at doses that would have totally inactivated the nonsporeformers studied in the pure culture experiments.

Table 2-E.1 summarizes the results to date on the effect of SER on microorganisms. Survival fractions for pure cultured spores and naturally occurring organisms are average values for the plateau regions of the survivor curves. Survival fractions for pure cultured nonsporeformers were at the 1×10^{-5} level (the limit of detection for the experimental technique employed) for all dose rate-temperature conditions at the $8,000$ ergs mm^{-2} dose level (also at $6,000$ ergs mm^{-2} for 1.0 sun tests).

The computerized statistical analysis of pure cultured organism data was initiated. The program is designed to reduce the data for each experimental run and calculate the mean survival fraction and standard deviation.

2.5.4 Future Activities

Pure cultured organism data analysis will be completed and a paper prepared for the open literature. Studies will be continued to define the SER resistance of naturally occurring organisms. In addition, experiments will be designed to study the effect of viable and nonviable particulate shielding on survival, and the resistance to SER in terms of microcolony forming ability.

2.5.5 Presentations

Wardle, M. D., "Effect of Solar Electromagnetic Radiation on Microorganisms," presented at the 14th NASA Spacecraft Sterilization Technology Seminar, Cocoa Beach, Florida, August, 1975.

Table 2. E-1. Summary of Approximate Survival Fractions of Test Populations Exposed to SER

| Dose Rate, Temperature | Pure Cultured | | Naturally Occurring ^b |
|--|--------------------|-----------------------------|-------------------------------------|
| | Spore ^a | Nonsporeformer ^a | |
| 0.1 SUN, -125°C | 1×10^{-3} | 1×10^{-5} | — |
| 0.5 SUN, -15°C | 2×10^{-4} | 1×10^{-5} | 7×10^{-2} (65°C) |
| 1.0 SUN, 70°C | 5×10^{-5} | 1×10^{-5} | — |
| ^a 1×10^{-5} = Limit of detection ^b 1×10^{-4} = Limit of detection | | | |

SECTION III

POST LAUNCH RECONTAMINATION STUDIES
(NASA No. 193-58-62-03)

Contents

Title and Related Personnel

para. 3.1

POST LAUNCH RECONTAMINATION
STUDIES

Cognizance: J. Barengoltz

Associate
Personnel: I. Ickovits (Bionetics)

3.1 POST LAUNCH RECONTAMINATION STUDIES

3.1.1 Introduction

The objective of the task is the development of an analytical technique for the evaluation of the probability of the relocation of particles from nonsterile to sterile areas on a spacecraft. The recontamination process is important for all multiple missions with separate microbiological burden allocations for various major spacecraft systems, and critical for life detection experiments that risk contamination from nonsterile components.

The approach has been to study the effects of typical mission environments on the redistribution of particles on spacecraft surfaces both analytically and experimentally. This study consists of three logical components, which have been reflected in the effort: (1) particle adhesion, (2) dynamic release mechanisms, and (3) particle transport. The effort in particle adhesion has been principally a particle release experiment, together with analytical work and attempts to correlate other data found in the literature and elsewhere. Under dynamic release mechanisms, meteoroid impact and pyro firing have been modeled. The particle transport activity is an analytical effort which includes the development of codes for spacecraft geometry and orientation, forces acting on released particles, and trajectory.

Finally all of these components were assembled into an operational, integrated computer code. For a demonstration calculation with this computer code, a geometrical model based on a Viking-type spacecraft and the spaceflight phase between Earth orbit and Mars encounter were chosen.

3.1.2 Significant Accomplishments

During this report period a comprehensive report on the analysis, computer code, and sample calculation for the spaceflight phase of a dual-element spacecraft has been completed (Ref. 1). The results of the sample calculation and some comparisons of analytical and experimental values may be found in previous semi-annual reports (Ref. 2 and 3).

The direction of the effort has changed to address the recontamination hazard for shuttle (STS) - launched planetary spacecraft. Areas of concern where the current analyses are inadequate or inappropriate to these missions are being identified. New work in the areas of particulate adhesion in partial vacuum, abrasive particulate generation, dynamic sun-shade geometries, atmospheric drag, and spacecraft electric field is being planned or underway.

3.1.3 Future Activities

The current effort, just begun, to adapt and improve the analytical tools developed previously in order to apply them to shuttle missions will continue. A significant activity in the particulate transport field will be undertaken to accomplish this goal. Some analyses of particulate adhesion and dynamic release environments particular to shuttle will also be performed.

3.1.4 Presentations

Barengoltz, J., "Post Launch Recontamination Studies," presented at NASA Spacecraft Sterilization Technology Seminar, Cocoa Beach, Florida, August, 1975.

3.1.5 References

1. Barengoltz, J. and Edgars, D., "The Relocation of Particulate Contamination during Spaceflight," NASA TM 33-737, Jet Propulsion Laboratory, Pasadena, California (1975).
2. Planetary Quarantine Semi-Annual Review, Supporting Research and Technology, 1 July - 31 December 1974, JPL Doc. 900-701, p. 3-1 (1975).
3. Planetary Quarantine Semi-Annual Review, Space Research and Technology, 1 July - December 1973, JPL Doc. 900-655, p. 3-1 (1974).

SECTION IV

SPACECRAFT CLEANING AND DECONTAMINATION TECHNIQUES
(NASA No. 193-58-63-02)ContentsTitle and Related PersonnelSubtask A
para. 4.1PHYSICAL REMOVAL OF SPACECRAFT
MICROBIAL BURDEN

Cognizance: H. Schneider

Associate
Personnel: W. Neiderheiser (Bionetics)Subtask B
para. 4.2EVALUATION OF PLASMA CLEANING AND
DECONTAMINATION TECHNIQUESCognizance: S. Fraser (Boeing)
D. TaylorAssociate
Personnel: R. Olson (Boeing)
W. Leavens (Boeing)Subtask C
para. 4.3EVALUATION OF VACUUM/HEAT
STERILIZATION

Cognizance: A. Irons

Associate
Personnel: J. Barengoltz
C. Hagen (Bionetics)Subtask D
para. 4.4

CLEAN TRANSFER SYSTEMS

Cognizance: A. Irons

Associate
Personnel: M. Peelgren
E. CuddihySubtask E
para. 4.5

ADVANCED BIODETECTION METHODS

Cognizance: J. Jacobs

Associate
Personnel: L. Goforth
F. Morelli (Bionetics)

4.1 PHYSICAL REMOVAL OF SPACECRAFT MICROBIAL BURDEN

4.1.1 Subtask A Introduction

Present planetary quarantine constraints for flyby and orbiter vehicles require maintaining the microbial burden on the spacecraft below a certain critical level. State-of-the-art clean room facilities and contamination control techniques do not assure that this critical level can be maintained throughout necessary assembly and test operations.

Previous activities under this task concentrated on the study of vacuum techniques with and without the use of a brush, and on steady state as well as pulse blow cleaning techniques using extra dry, high-purity nitrogen. Test devices were developed that would allow for the simulation and evaluation of these techniques under controllable conditions with an observation of the behavior of the test particulates under a 100X microscope. Studies to establish the mechanical properties of candidate brush-materials were conducted to the JPL materials test laboratory. As described in detail in JPL documents 900-597, 900-636, 900-675, and 900-701 the following findings and deductions were made:

- 1) Conventional vacuum brushes efficiently detach particles of the smallest discernible sizes ($2-3\text{ }\mu\text{m}$) but the flow velocities near the surface are too low to entrain and to transport detached particles into the vacuum system. Consequently, the bristles quickly become saturated with particles, and the brushes become a shedding source (rather than a cleaning tool) if not cleaned frequently between uses. To eliminate this problem, further developments would have to consider improved entrainment techniques (i. e., less flow resistance across the brush) and self cleaning capability.
- 2) Vacuum flow alone efficiently detaches and removes particles larger than $10\text{ }\mu\text{m}$ if, the surface is dry and flow velocities are near critical (choked flow). A nozzle stand-off distance from the surface on the order of $150-200\text{ }\mu\text{m}$ is required. The removal efficiency drops off sharply at larger stand-off distances.

Vacuum cleaning, therefore, has only a potential for the cleaning and sampling of smooth and even surfaces.

- 3) Blow cleaning by means of a 45 degree jet efficiently removes particles greater than 5 μm from normally dry (i. e., <60% RH) surfaces at a system pressure of 30 psig. Although this pressure complies with safety regulations, the resulting impingement will not be compatible with many spacecraft surfaces.
- 4) A periodic positive blocking of the jet noticeably enhances removal efficiency by 20-30% where otherwise relatively low removal efficiencies <50% are achieved. Best results are obtained at pulse frequencies below 50 Hz and pressures producing just near critical flow across the nozzle. This method has attractive points for use in devices specifically designed for the spotcleaning of semi-occluded areas that cannot be reached otherwise. (See Rep. 900-675, Fig. 4-A-21.)
- 5) Jet deflection (rather than blocking) induced by blowing against wedges and over oscillating rods did not improve detachment. Efficient (nearly 100%) removal of particulates of all sizes, however, could be produced by feeding isopropyl alcohol through a hollow rod blown across at pressures between 1 and 3 psig to induce vibration. The method is not suited for large area cleaning but seems to have a potential for spotcleaning and biological sampling (see para. 4.1.3.3).
- 6) The application of ultrasonic techniques (Hartmann generator) in a synergistic mode with flow did not show any improvement over what could be accomplished with vacuum flow alone. Probable causes for this are insufficient power output of the existing apparatus as well as physical limitations, which to establish was not within the scope of the task, (expansive test equipment required).
- 7) The cleaning of surfaces temporarily moistened by inducing condensation prior to test was very inefficient under all the conditions

tested, which demonstrates the absolute need for drying before cleaning can be commenced, if an accidental moisture condensation has taken place.

- 8) Natural sable hair (presently in use for spacecraft cleaning) has a relatively low fatigue resistance and tends to shred with continuous use. Dupont Felor fiber was found to have the most acceptable properties for use in motorized cleaning brushes.
- 9) The capability of Felor fiber to pick up and to retain 5 to 25 μm particles increases by a factor of 2 if the fiber is mechanically flagged (rough surface), and by a factor of up to 4 if flagged and charged. Static charge, however, affects only the number of sweeps needed to accomplish efficient removal. 100% removal is achieved in 4 sweeps with charged brushes. Four (4) to 6 more sweeps are required to achieve 100% removal with uncharged brushes. On motorized brushes producing a large number of sweeps per unit time, allowing the fibers to become charged does not contribute to the efficiency of the device. Grounding the fibers, therefore, appears to be the right approach from the safety standpoint.
- 10) Brush fiber cleanliness can be restored by one single sweep over a vacuum grid. Striker-bar cleaning of the brush in vacuum flow requires 4 to 5 repeats to achieve total removal of the adhering particulates. In motorized brushes this can be accomplished within a 30° or less rotation after sweeping the contaminated surface.
- 11) The amounts of particles shed from the brush before the fibers reach the cleaning station (vacuum grid or striker-bar), are on the order of 10 to 30%. Provisions have to be made that these particles are also entrained into the flow and are transported into the vacuum system, i. e., ample flow between the fiber lift-off point and the vacuum grid (or striker bars) is necessary to prevent particles from falling back onto the surface.

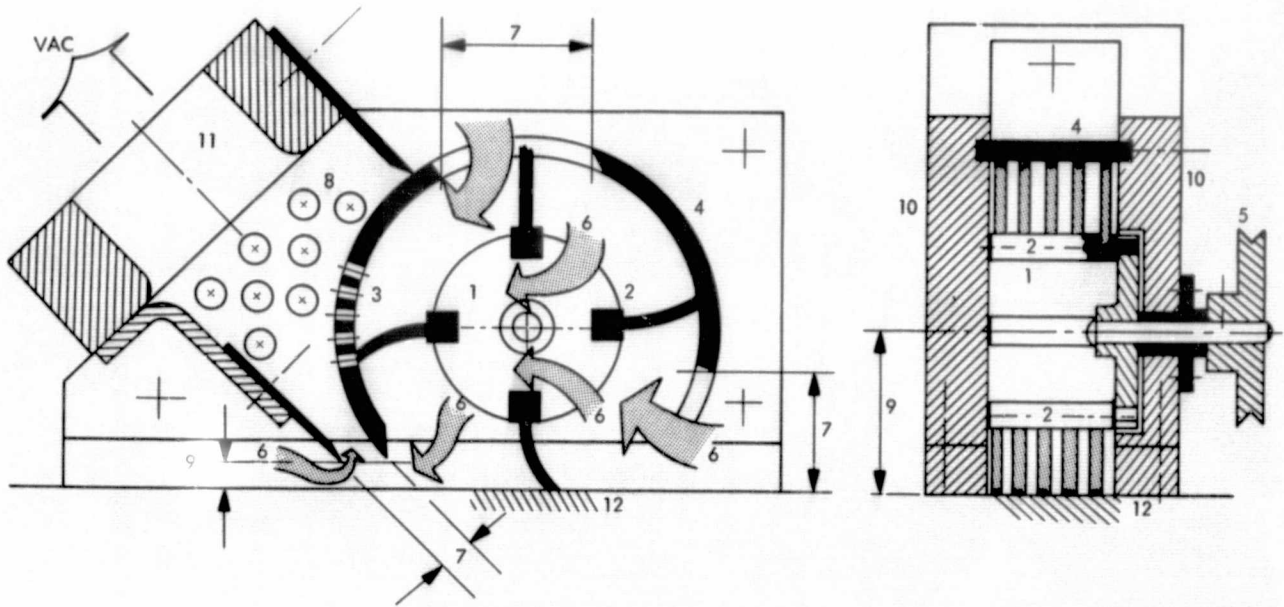
4.1.2 Rotary Brush Tests

4.1.2.1 Approach. The primary objective during this reporting period was to demonstrate the biological cleaning capability of an experimental motorized self-cleaning brush design based upon the findings and recommendations derived from previous tests conducted under this task. The test was conducted in two phases. In Phase I 50 x 50 mm optical glass slides were used as sample surface with the objective of establishing the effect of a repeated use of the device on its own state of cleanliness, i. e., background. In Phase II 23 x 50 mm stainless steel coupons roughened with #6 sandpaper were used as sample surface, with the objective to establish the effect of cross-sweeping on the cleaning efficiency of the device.

The test samples were exposed to the fallout of an uncontrolled shop area for the period of 1 to 3 weeks prior to test. Background and control samples were sterilized in dry heat at 130°C for a period of 24 hrs. The tests were conducted on a laminar flow bench in an air conditioned laboratory room at 22° C and 58% RH. The test apparatus was submersed in 90% isopropyl alcohol for 30 min and was air dried at 55°C for a period of 45 min the day before the test. The apparatus exterior surfaces were wiped with 90% isopropyl alcohol shortly prior to each phase of the test. Smocks, caps and gloves were worn by the operating personnel.

For assay the contaminated samples were sonicated in 30 ml peptone each for the duration of 12 min. 1 ml out of each 30 were then pipetted into 6 individual petri dishes and were plated in even parts at the original and at 1/10 of the original concentration (as a precaution for high count). The sterile controls were also sonicated in 30 ml peptone each, but 20 ml out of 30 were pipetted into Renier dishes for plating (because of the anticipated low counts). Agar was poured and incubation conducted at 32°C.

4.1.2.2 Apparatus Description. Figure 4-A.1 shows a schematic sketch of the assembled test device. The brush rotor (1) was pulley driven at a constant speed by means of a small (65 W) induction motor. Four V-shaped flagged Felor nylon computer drum brushes of the type ADP-10 manufactured by the Anchor Brush Co., Aurora, Ill. were used as brush elements (2). The



MAJOR DESIGN FEATURES:

- 1 - FREEBREEZING CAGE TYPE ROTOR
- 2 - EXCHANGEABLE BRUSH ELEMENTS
- 3 - ADJUSTABLE VACUUM GRID SEGMENT
- 4 - CHARGE OR GROUNDING SHOE
- 5 - EXCHANGEABLE PULLEYS
- 6 - ROTOR CROSS FLOW

- 7 - VARIABLE FLOW CONTROLLING AREAS
- 8 - BYPASS ORIFICES, PLUGGED OR OPEN
- 9 - VARIABLE STAND OFF FROM SURFACE
- 10 - PLEXIGLAS SIDEWALLS (FOR STROBE OBSERVATION)
- 11 - VAC HOSE TO FACILITY VAC SYSTEM
- 12 - USE WITH BASE OF BRUSH TEST APPARATUS

Fig. 4-A.1. Experimental rotary brush, 25 mm (1 inch) sweep width

elements could be radially adjusted by means of four eccentrics, (visible in Fig. 4-A.2) for desirable hairbending, when sweeping over the sample surface (12) or over the vacuum grid (3), which was established by means of a strobe light. The side walls of the device were made from plexiglass for this particular purpose. The baseplate used in previous linear sweep tests was applied to guide the test device and to position the samples (see Fig. 4-A.3).

For the discussed tests the following specifications did apply:

| | |
|-------------------------------|------------|
| Rotor Speed | 830 rpm |
| Fiber tip diameter (straight) | 72 mm |
| Working Width | 25 mm |
| Max. Fiber deflection | 30 degrees |
| Vacuum Nozzle Standoff (9) | 8 mm |

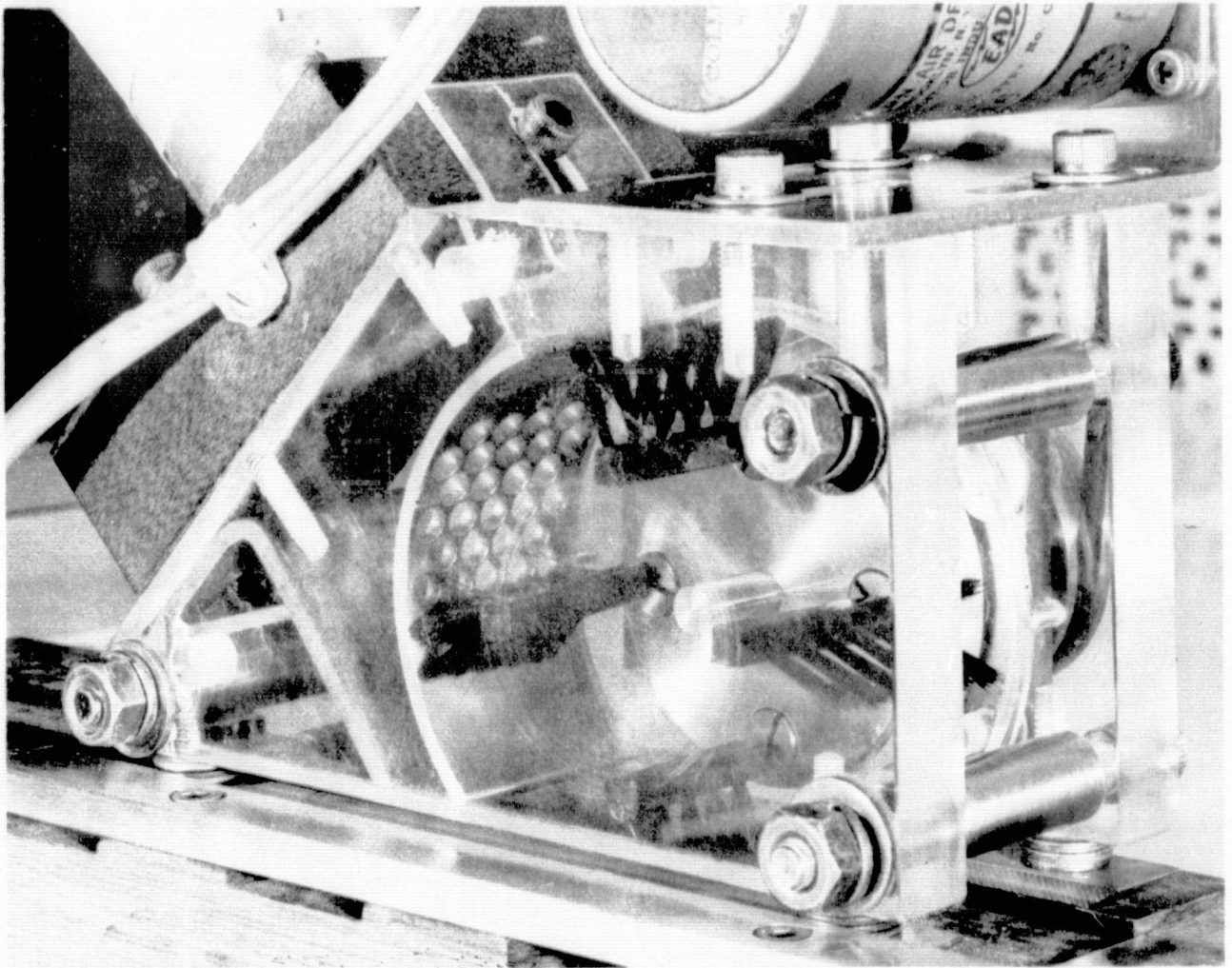


Fig. 4-A.2. Experimental self cleaning brush. A view of the rotor, brush elements, vacuum grid and nozzle

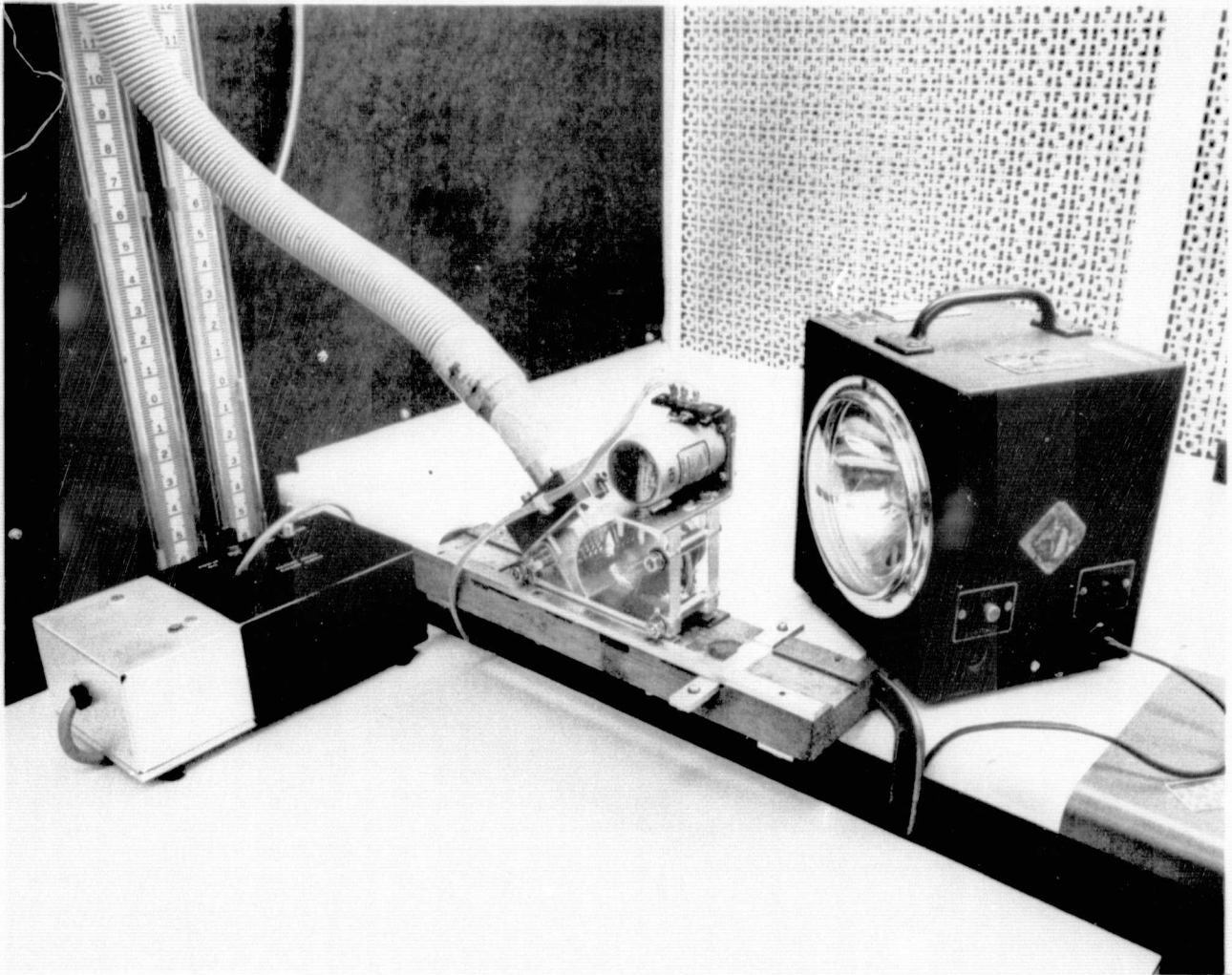


Fig. 4-A.3. Test set up. Rotary brush placed on baseplate providing for guidance and sample alignment. Strobe light for observation of fiber dynamics and adjustment

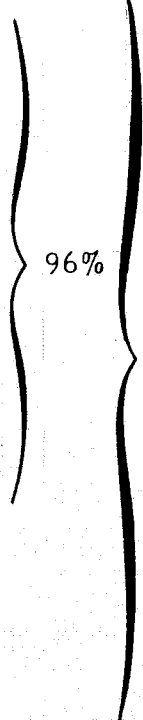









| | |
|---------------------------|-----------|
| Vacuum Nozzle Opening (7) | 8 mm |
| Bypass orifices (8) | Closed |
| Charge Shoe (4) | Removed |
| Vacuum Average Pressure | 150 mm Hg |

4.1.2.3 Accomplishments. The results obtained from a total of 50 tests are summarized in Tables 4-A.1 and 4-A.2. As mentioned earlier, the objective of the Phase I tests was to establish the test background and the self cleaning capability of the brush.

**Table 4-A.1. Phase I Rotary Brush Evaluation Test with
Facility Fallout on Optical Glass (Test sequence
indicated by arrows)**

| Procedure | Specimens | Number of Viable Organisms Present After Indicated Treatment (Average from 4 determinations each) | | | Remarks |
|---|------------------------------|--|-----|------|---|
| | | 1 | 2 | 3 | |
| Direct Transfer From Fall-out Tray into Assay Container | 1 Sterile Controls | 0.3 | | | 4 } AV. 110.9 } 86% AV Removal Effi- ciency |
| | 2 Contaminated Samples | 94.9 | | | |
| Samples Placed on Apparatus and Trans- ferred Into Assay Con- tainer Without Test | 3 Sterile Controls | 0.5 | | | |
| | 4 Contaminated Samples | 126.9 | | | |
| Cleaning with Three Over- lapping Paral- lel Sweeps (as shown in Fig. 4-A.4(A)) | 5 Sterile Controls | 1.3 | 1.3 | 3.0 | AV. 15.3 |
| | 6 Contaminated Samples | 20.8 | 8.9 | 16.3 | |

Table 4-A.2. Phase II Rotary Brush Evaluation Test
with Facility Fallout on Roughened Stainless Steel
(Test sequence indicated by arrows)

| Procedure | Specimens | Number of Viable Organisms Present After Indicated Treatment (Average from 4 determinations each) | | | | Remarks | |
|--|-------------------------------|--|--|--|--|---|-----|
| Samples Placed on Apparatus and Transferred into Assay Container without Test | 1 Sterile Controls | 1 0 | 2 | 3 | 4 | 5 Removal Efficiency  | |
| | 2 Contaminated Samples |  50.1 | | | | | |
| One Lengthwise Sweep Over Surface* | 3 Sterile Controls |  1.8 |  2.0 |  0.3 | 0.8 | | 96% |
| Two Lengthwise Sweeps Over Same Surface* | 4 Contaminated Samples |  1.7 |  0.8 |  2.4 | | | 99% |
| One Lengthwise Sweep Followed by Three Overlapping Lateral Sweeps Over Same Surface* | 5 Contaminated Samples | |  0.8 | |  0.3 | | |
| * See Fig. 4-A.4 (B). | | | | | | | |

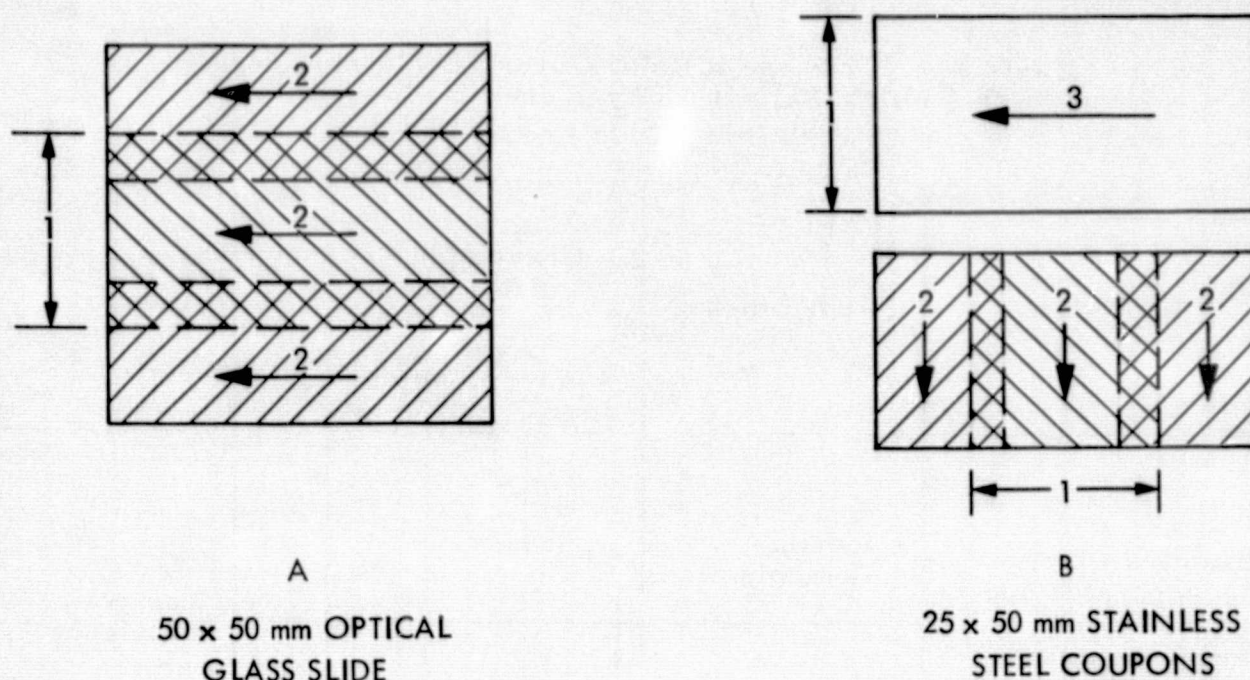


Fig. 4-A.4. Schematic of test sweep pattern
 1 Brush sweep width
 2 Overlapping parallel sweeps
 3 Single or multiple sweeps over same surface

As can be seen from Table 4-A.1, Col. 1, line 1 and 3, the contamination introduced due to handling is negligibly small ($<1/2\%$) if compared to the contamination found in the exposed test samples, (line 2 and 4, col. 1). It appeared to be reasonable, therefore, to combine the line 2 and 3 data in one average of 110.9 to serve as a baseline for cleaning efficiency determinations. With a total sample surface of 25 cm^2 , this represents a microbial burden on the order of 4.5×10^4 per m^2 , which, for example, is an order of the microbial burden encapsulation redline for spacecrafts operating in the vicinity of the planet Mars (fly-by or orbiting).

The sweeps over the sterile controls (line 5, col. 1, 2, 3) indicate a slight contamination of the sample surface due to brush contact, which appears to be progressive. But, being small (1 to 3%) if compared to the exposed test samples, this is still within the randomness that has to be anticipated. The removal efficiency obtained from a total of 12 tests is on the order of 86% (col. 4).

The objective of the Phase II tests (Table 4-A.2), as mentioned earlier, was to test cleaning efficiency on a rough surface, and to establish whether cross-sweeping is more efficient, if compared to sweeping twice over the same surface in one direction.

The test background (line 1), was practically zero and the average count of 50.1 (line 2) obtained from 4 test samples was used as a baseline for cleaning efficiency determinations. With a sample surface of 12.5 cm^2 this represents a microbial burden of 4×10^4 per m^2 , which is on the same order as obtained for the glass slides used in Phase I tests.

The sweeps over the sterile controls between tests (line 3, col. 1 through 4) indicate a slight contamination of the brush due to use which does not seem to be progressive in nature. This basically demonstrates the self cleaning capability designed into the device. The linear sweep efficiency obtained from 8 tests averaged to 96%, 10% higher than the efficiencies obtained during Phase I tests. This is due to the fact that each surface was swept twice (see 900-701, Fig. 4-A.13). Also, detachment forces tend to be lower on rough surfaces where moisture is a contributing factor to particle adhesion.

There is no doubt that cross-sweeping is the most efficient method (line 5, col. 2 and 4) producing an almost total removal of all viable particles on the surface. The 99% removal efficiency indicated in col. 5 represents an average of 8 tests.

4.1.2.4 Summary Conclusions and Recommendations. The tests conducted basically prove that a self-cleaning rotary vacuum brush that accomplishes an efficient biological cleaning of typical spacecraft surfaces can be developed. The experimental device tested is not necessarily representative of an optimum utilization of the chosen concept, which to establish will require a certain further development and field tests with prototype devices. At this point, the following general recommendations are given:

- a) For the first prototype the design should comply with the specifications listed in Para. 4.1.2.2, except where the indicated 25 mm width is concerned. A working width on the order of 100 mm appears to be feasible for handheld brushes. For brushes having positive standoff controls that assure plane parallel movement, a

larger working width up to 300 mm appears to be feasible, which will be limited by the rotor structural strength and dynamics.

- b) The flow cross-sectional areas of the vacuum nozzle, the vacuum grid and the hose-connection total inlet area (if more than one vacuum hose is used) should be proportional to the width of the experimental device. The flow controlling dimensions of the nozzle as well as the surface standoff (Fig. 4-A.1), should be adjustable within $\pm 50\%$ of the dimensions indicated in Para. 4.1.2.2 for the experimental device.
- c) To assure proper fiber contact with the surface to be cleaned and with the vacuum grid, the rotor design must provide for ample cross-sectional area for the flow to stream around the fiber tufts without causing a lift-off from the surface, tuft reversals or entanglements. The cage-type rotor concept which allows the flow to pass through the rotor center satisfies these requirements for practically two dimensional flow as existent for wide rotors. The experimental device simulated a wide rotor aerodynamically by providing solid side walls. On an actually working model the side walls can be perforated or be completely omitted, except for the structures necessary to provide for the rotor bearings mounts.
- d) For prototype designs, the rotor drive mechanism should provide for a continuous or incremental variation of the rotor speed from approximately 500 to 1500 rpm. For field application a fixed rpm induction motor pulley drive appears to be the best approach from the reliability standpoint. A motor having 25 to 30 watts rated input per cm-rotor width is recommended for continuous operation.
- e) For practical purposes the design should consist of the following major modules (see Fig. 4-A.1):
 - 1) A basic body incorporating the vacuum plenum (8), the nozzle (7), the vacuum grid (3), the side walls (10) and the rotor bearing mounts. This structure should be compatible with rigorous sterilization and decontamination procedures.

- 2) A quick-exchange rotor assembly consisting of a sterilizable rotor hub (1) including bearings and exchangeable brush elements (2) compatible with a complete submersion in isopropyl alcohol.*
 - 3) A removable drive mechanism consisting of motor, pulley drive and/or gears, compatible with external wiping with isopropyl alcohol.
- f) The device should be advanced on the surface in a direction normal to the rotor axis, not exceeding 3 cm per second. Since the sweep velocity of the fibers is on the order of 3 m per second, the device can be worked in either direction, with or against the rotor sweep. Sweeping towards the operator (i. e., pulling the device) is recommended to minimize recontamination of the cleaned area. Succeeding sweeps under 60 to 90° crosswise to each other appears to be the best method from the removal efficiency, and from the recontamination standpoint.

4.1.3 Particle Adhesion Test

A second objective of the discussed reporting period was to generate particle adhesion data applicable to the post launch recontamination study conducted under Task No. 193-58-62-03. The tests conducted during this reporting period concentrated mainly on establishing the effects of detachment angle and typical surface conditions (dry, fogged) on the detachment force under atmosphere conditions.**

*The bearing design will depend on the application of the device. For operations where the presence of oil is not a major problem, sleeve or ball bearings requiring small amounts of sterilized low viscosity oil should be adequate. At operations where stringent organic cleanliness requirements are in existence, continuous dry running capability will be mandatory for the bearings.

**The test apparatus has also the capability to conduct tests under vacuum conditions.

4.1.3.1 Approach. To accomplish this 5 x 10 mm samples cut from standard bio-slides were seeded with glass beads of selected size ranges and were spun at room temperature in an air conditioned lourdes centrifuge in predetermined speed increments up to a maximum detachment force of 33,000 G's.* Always two samples were spun at a time in opposing positions of the centrifuge. Number counts were taken from two separate 1/2 x 1/2 mm areas of each individual sample under a 100 power microscope after seeding and between tests. Size distribution determinations were made from each sample after seeding, and for certain selected G-levels, also after test.

To simulate surface moisture condensation (as it may potentially occur during spacecraft operations due to facility systems failure) the seeded samples were placed in a refrigerator for a period of 15 minutes to induce visible fogging when returned to the room atmosphere. The fogging, then, would disappear after a few minutes, and the samples were spun 15 to 20 minutes later.

4.1.3.2 Apparatus Description. Figure 4-A.5 shows one of the individual 5 x 10 mm glass samples mounted (glued) on a cylindrical holder, positioned for counting. The notched circular disc is a positioning device that assures counting from one and the same area prior to and after test. For the test, the sample-holders (cylinders) were inserted under a prescribed angle into a holding rack, the seeded side facing radially out, as shown in Fig. 4-A.6 and 4-A.7. Two racks at a time, then, were inserted in opposite positions of the centrifuge rotor, as shown in Fig. 4-A.8.

During the discussed tests the rotor buckets would remain open, but the top opening was closed to avoid disturbance of the samples due to turbulence. The centrifuge cooling system could be adjusted to maintain the prevailing room temperature during the test. The device shown in Fig. 4-A.9 allows to seal the rotor buckets through the cover flange of a high vacuum chamber.

* $1G = 9.81 \text{ ms}^{-2}$.

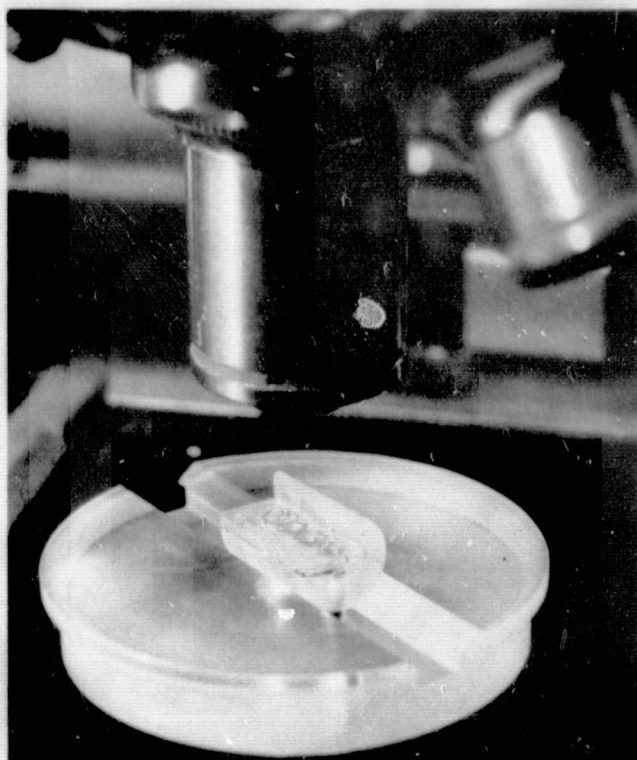


Fig. 4-A.5. Sample holder positioned for counting under 100 X microscope

4.1.3.3 Accomplishments. At the time of this report the atmospheric portion of the test has been completed. An evaluation pertaining to adhesive properties and numerical size/force relations is in progress. In the following, the test data as available at this point will be presented and be briefly discussed.

Summarized in Table 4-A.3 are the environmental conditions in existence during the time of each individual test, and size-related parametric data of the test material as seeded on the sample surface. Figs. 4-A.10 and 4-A.11 show typical size distributions as existent on the samples prior to centrifugation. Figure 4-A.12 shows the removal fractions obtained from 2 counts each taken after centrifugation to the indicated G-level. For the 5-10, 20-25 μm size ranges having a size average of 7-8 and 20-22 μm (see Table 3) it is quite obvious that a normal-to-surface (90°) detachment requires about twice the force necessary for a surface-parallel detachment. For the $\leq 5 \mu\text{m}$ sizes this is not quite conclusive. In both circumstances substantially high G-loads on the order of several thousand G's are necessary to affect a noticeable removal of particles of the considered size ranges.

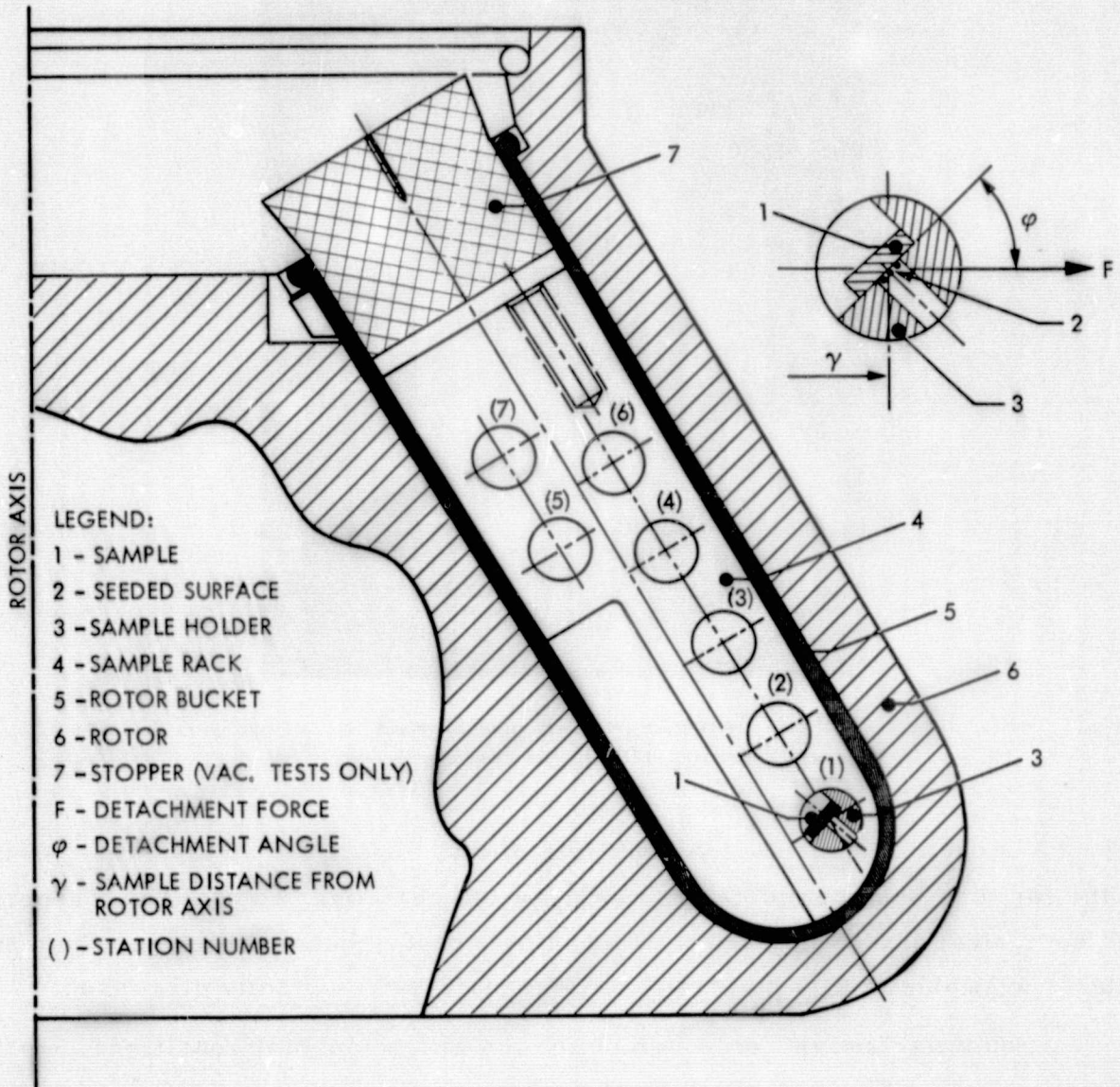


Fig. 4-A.6. Sample installation and nomenclature

Figure 4-A.13 compares identical data obtained from tests with dry and with fogged samples, for a normal to surface detachment. It can be seen that the presence of residual moisture, such as caused by a previously occurred surface condensation, increases the detachment force by one order of one magnitude and more, if compared to the normally dry ($RH \leq 60\%$) surface.

At levels below 1000 G's the data obtained became inconsistent probably because vibratory forces generated by the centrifuge itself became

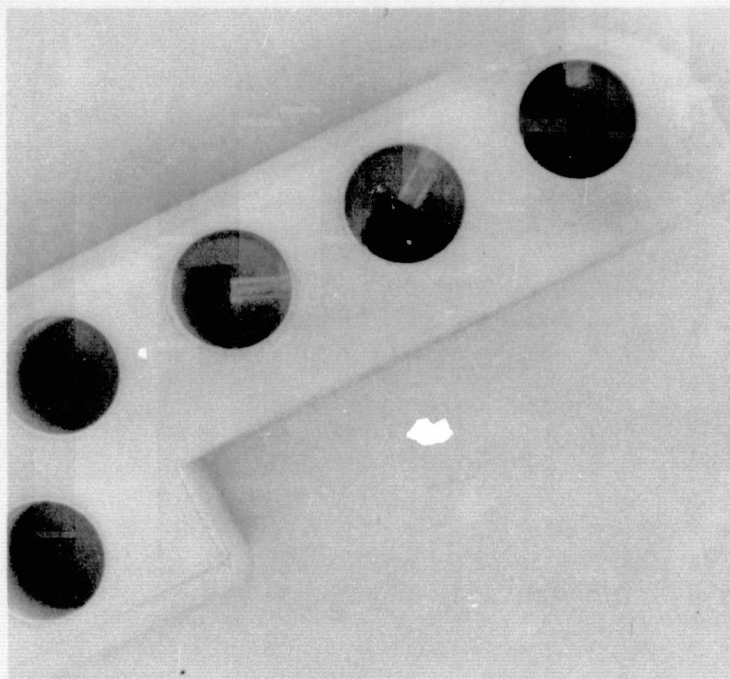


Fig. 4-A.7. Sample installation in positioning rack

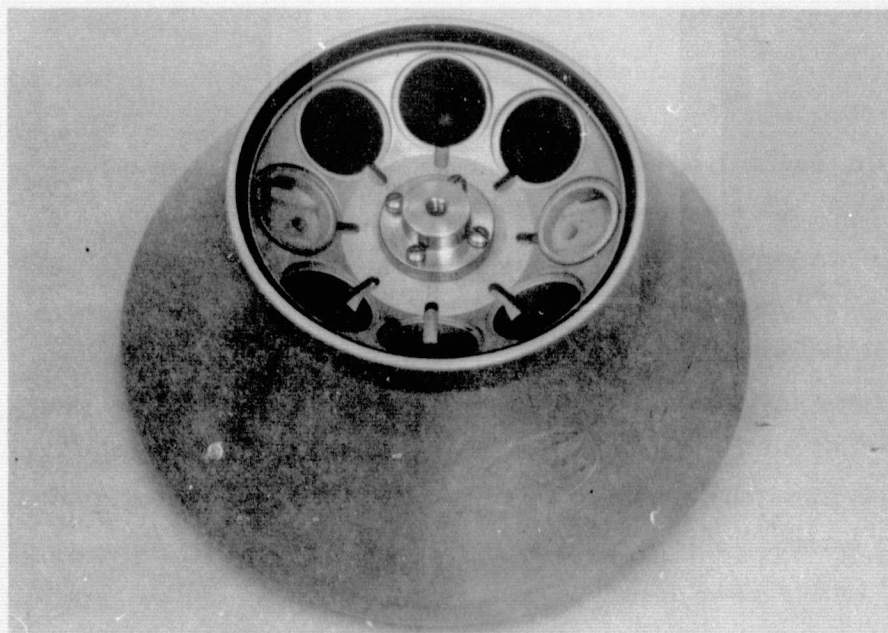


Fig. 4-A.8. Installation of sample racks in centrifuge rotor

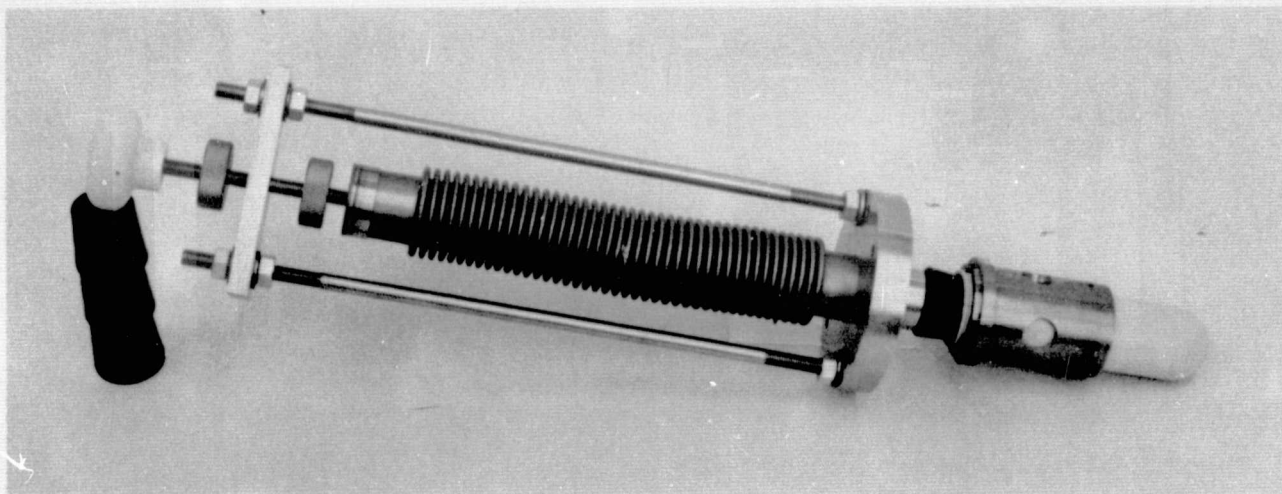


Fig. 4-A.9. Device to accomplish sealing of sample container through wall of vacuum chamber

more pronounced, relative to the centrifugal force acting on the particles. This made it difficult to establish threshold data, i. e., G-loads at which the particles actually start moving. The data points below 1000 G's, therefore, were omitted in this report pending additional tests to obtain more data points for a statistical treatment of this particular region.

All of the above discussed data were derived from total counts comprising all discernible sizes present on the samples. Being of particular interest for the evaluation of particle detachment due to flow, size distributions were established for a few selected tests with particles $<10\text{ }\mu\text{m}$ on a dry surface with a parallel to surface application of the detachment force. As can be seen from Figure 14, detachment starts with the largest sizes, spreading fairly evenly over all sizes larger than the present mean, which decreases as the detachment force increases. In Fig. 4-A.15 it is interesting to note that an exponential relationship between the adhesive force (F) and the particle size parameters (d) according to $F = k; d^a$ prevails which will be further evaluated. Extrapolating towards the parameters of the original distribution ($G = 0$) of the test material indicates a detachment threshold on the order 2000 to 3000 G's for the considered test.

Plotted in Fig. 4-A.16 are the removal fractions for one μm size increments. It is very apparent from this plot that, with atmospheric moisture

Table 4-A.3. Test Parameters and Conditions. Centrifugation Tests w/Glass-Bead on Glass

| Surface Condx | Dry (According to RH Indicated) | | | | | | | | Intentionally Fogged | | | |
|--|---------------------------------|------|-------|---|-------------------------------|------|-------|---|-------------------------------|------|-------|---|
| Detachment Angle | In Surface Plane 0 degr. | | | | Normal to Surface 90 degr. | | | | Normal to Surface 90 degr. | | | |
| Nom. Size Range, μm As Indicated by Supplier | <5 | 5-10 | 20-25 | X | 0-5 | 5-10 | 20-25 | X | <5 | 5-10 | 20-25 | X |
| Initial Av. Size, μm^* | 4.07 | 8.84 | 20.50 | X | 3.50 | 8.53 | 22.30 | X | 2.64 | 7.08 | 22.03 | X |
| Initial Std. Deviation, μm^* | 1.70 | 2.40 | 3.70 | X | 1.47 | 2.15 | 3.90 | X | 1.60 | 2.97 | 4.05 | X |
| Temp, $^{\circ}\text{C}$ | 20.0 | 21.7 | 21.1 | X | 20.0 | 21.7 | 21.1 | X | 21.1 | 21.1 | 21.1 | X |
| RH, % | 58 | 50 | 56 | X | 58 | 50 | 56 | X | 58 | 50 | 58 | X |
| Pressure | Atm. | | | | | | | | | | | |
| *As seeded on sample surface. | | | | | | | | | | | | |

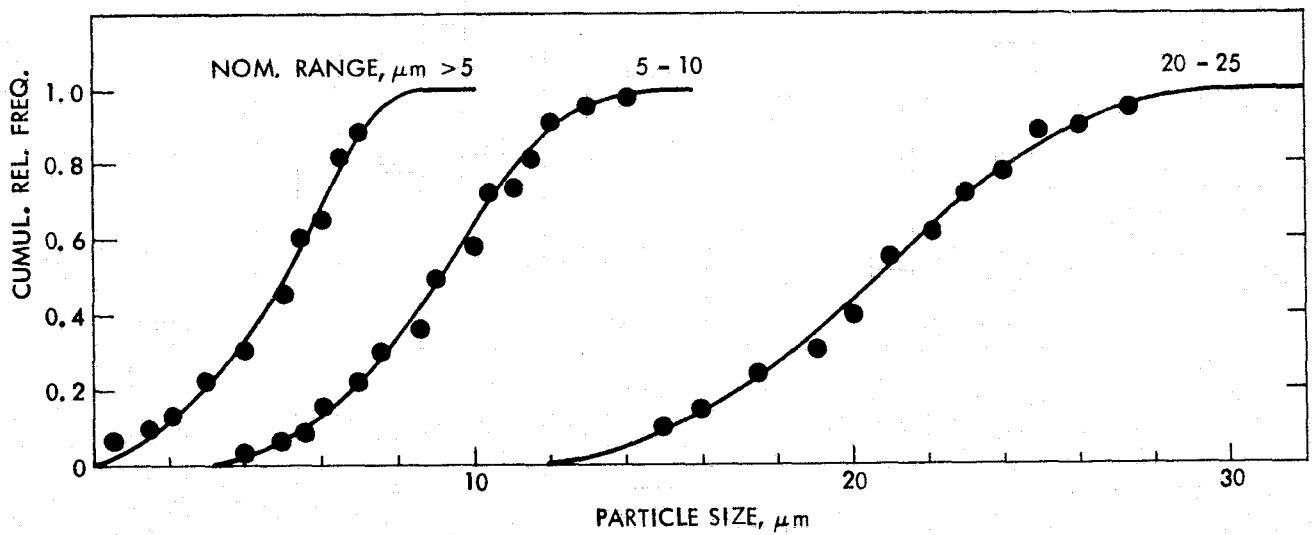
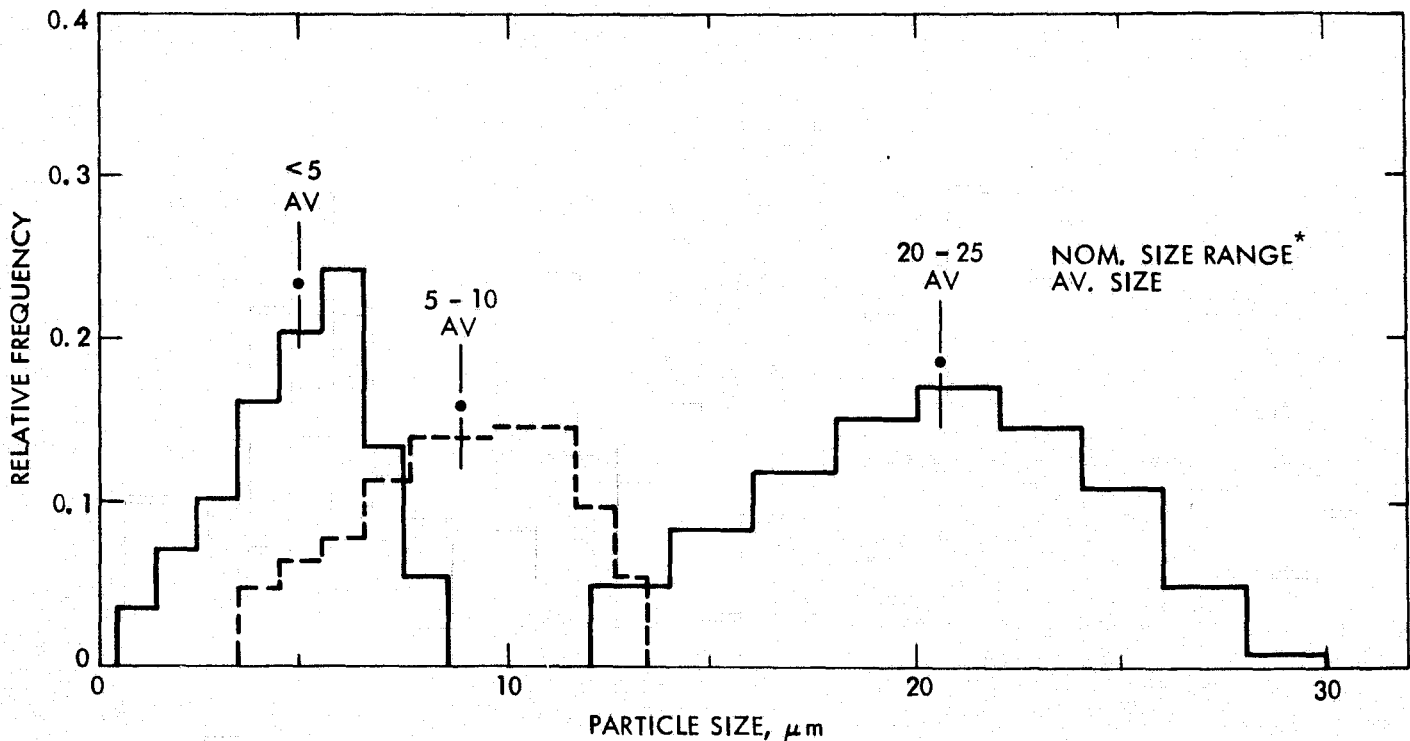


Fig. 4-A. 10. Typical cumulative counts taken from seeded samples prior to test



*As indicated by supplier.

Fig. 4-A. 11. Typical size distribution of seeded test material prior to test

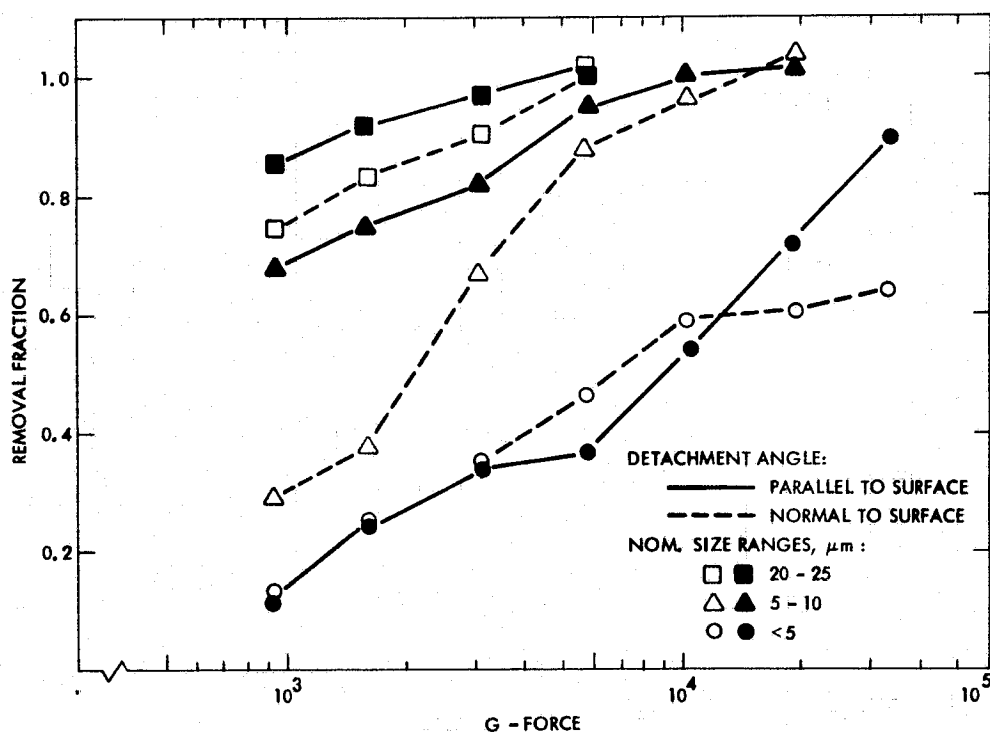


Fig. 4-A.12. Effect of size and force angle on particle detachment (glass beads on dry glass)

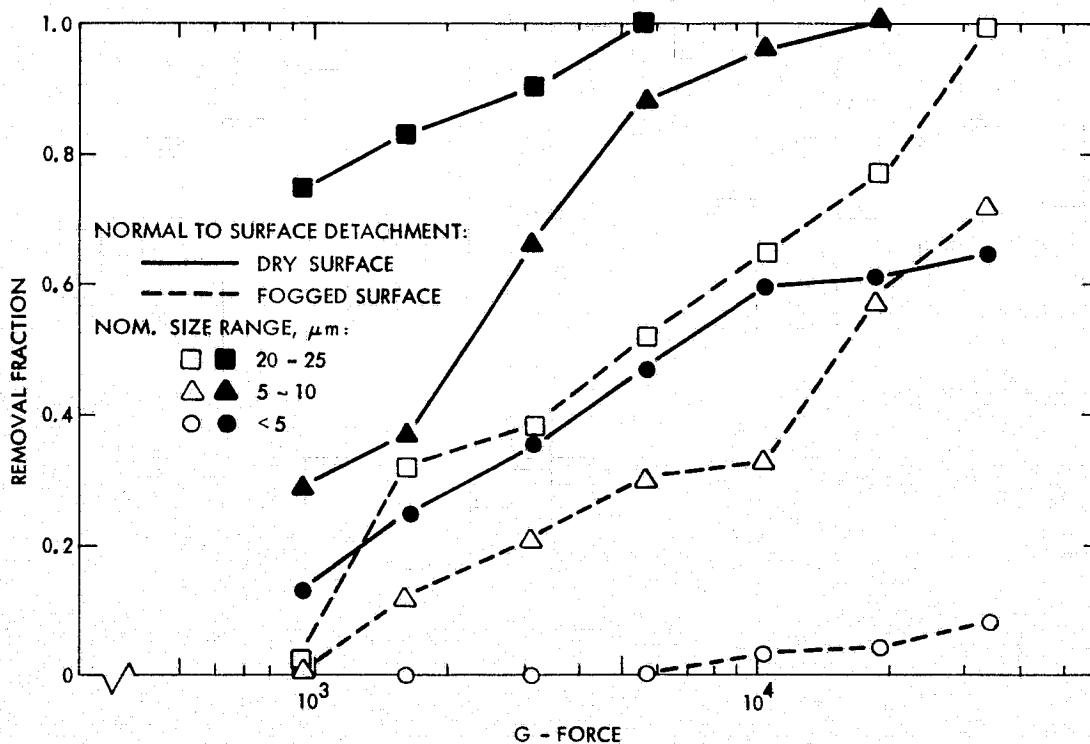


Fig. 4-A.13. Effect of size and surface conditions on particle detachment (glass beads on glass)

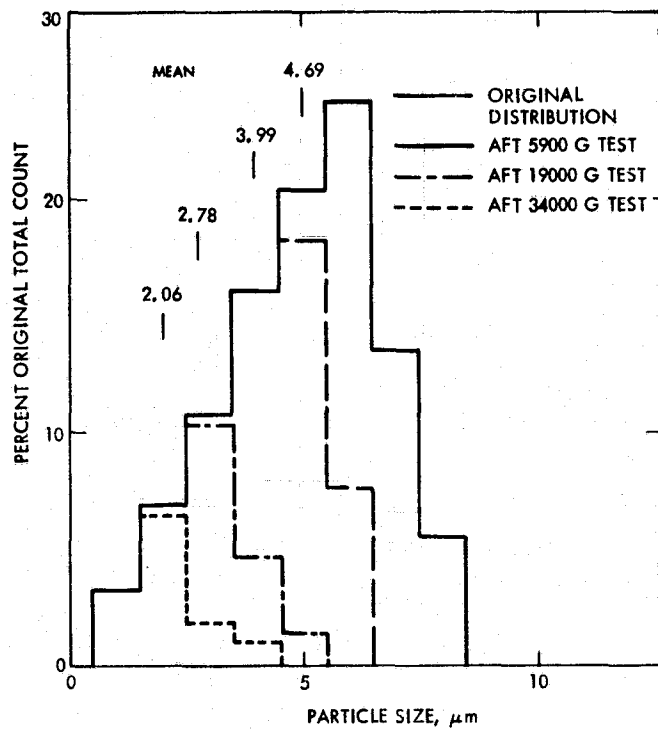


Fig. 4-A.14. Effect of G-Force on size distribution of material remaining on surface. Glass beads on dry glass parallel to surface detachment

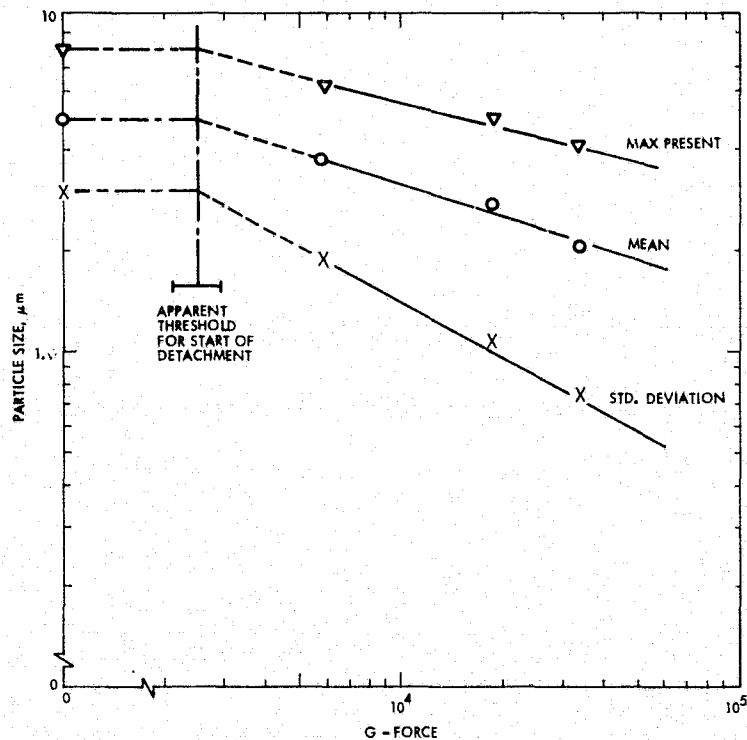


Fig. 4-A.15. Effect of G-force on size parameters. Glass beads on dry glass parallel to surface detachment

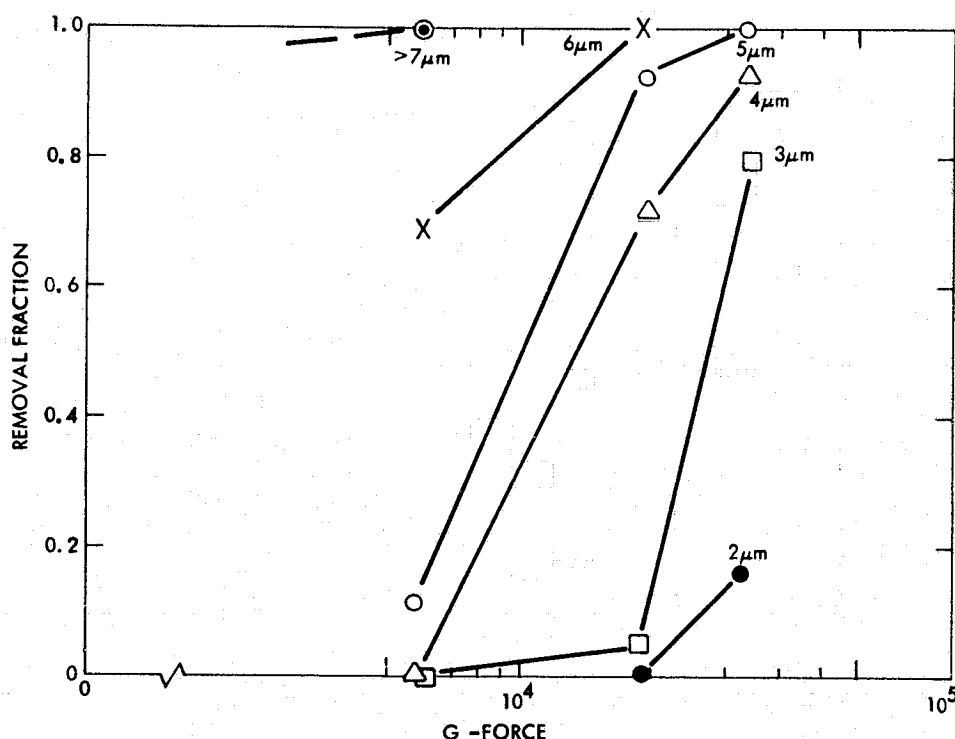


Fig. 4-A.16. Increment removal fractions vs G-load.
Glass beads on dry glass parallel to
surface detachment

present, the detachment of small particles ($<5 \mu\text{m}$) is only practicable by means of a physical force (such as a brush), or by application of a high density medium, such as a liquid flush.

4.1.4 Future Activities

4.1.4.1 Physical Removal of Biological Burden. A final report summarizing all work conducted under this task is being prepared. The report will also discuss the mechanics, the potentials and the limitations of the methods tested, utilizing particle adhesion data obtained from the atmospheric centrifugation tests described in Para. 4.1.2. According to the task objective, general guidelines for the design and use of biologically efficient particulate cleaning devices will be established. This will then conclude the work funded under this task. Additional effort will be required for the implementation of selected specific design approaches we believe are most suited to fill the needs for spacecraft cleaning.

4.1.4.2 Particle Adhesion Tests. Vacuum centrifugation tests with glass beads utilizing the apparatus described in Para. 4.1.3.2 will be commenced as soon as the vacuum chamber is available. The analytical evaluation of the test data is in progress, with the objective to support final evaluation of the removal efficiency experiments with flow, and to establish analytical tools for the assessment of the particulate removal probability from exterior (spacecraft and/or Shuttle) surfaces due to environmental effects, such as vibrations, shock, exterior aerodynamics and space vacuum.

4.1.4.3 Liquid Film Biological Sampling. Another look will be taken at a removal method described in Report 900-675, pages 4-16, 4-17, where a thin film of isopropyl alcohol was fanned onto the surface by means of an oscillating hypodermic needle. A new apparatus will be designed and fabricated that will simulate the oscillating film mechanics in a manner that has the potential for the biological sampling of surfaces. The concept envisioned at this time to accomplish this provides for a liquid jet aimed at the edge of a vibrating tongue, with the liquid (rather than the vacuum flow) being the primary driver. The vacuum flow will only be utilized to transport the detached organisms into a filter or a liquid trap. Cyclonic action rather than a filter will potentially be utilized to separate the organisms from the flow. The tests will be conducted under 100X with $<5\text{ }\mu\text{m}$ glass beads, and on biological fallout samples, and will be compared with swab-rinse and with Sandia probe sampling methods.

4.2 EVALUATION OF PLASMA CLEANING AND DECONTAMINATION TECHNIQUES

4.2.1 Subtask B Introduction

The objective of this task is to develop plasma sterilization technology that can be applied to problem areas in the decontamination and sterilization of spacecraft hardware.

4.2.2 Significant Accomplishments

All three phases of this task have been completed and the preparation of a final report is underway. The overall findings of the study can be summarized as follows:

- 1) Of the radio frequency generated gas plasmas studied, helium plasma was found to be the most effective in inactivating microbial populations.
- 2) Plasma produced at 0.2 mm Hg was most efficient as a lethal agent.
- 3) Energetic photons account for the lethality of plasma.
- 4) Plasma is an effective surface sterilant capable of killing microorganisms on surfaces of various geometric configurations.
- 5) Plasma gases are compatible with most spacecraft materials at normal sterilizing exposures.

4.2.3 Future Activities

A final report on this task will be issued in September, 1975.

4.2.4 Presentations

Fraser, S., "Evaluation of Plasma Cleaning and Decontamination Techniques," presented at the NASA Spacecraft Sterilization Seminar, Cocoa Beach, Florida, August, 1975.

Fraser, S., "Sterilization with RF-Generated Cold Plasma," presented at 75th annual meeting of the American Society for Microbiology, New York, N. Y., April, 1975.

4.3 EVALUATION OF VACUUM/HEAT STERILIZATION

4.3.1 Subtask C Introduction

The objective of this subtask is to determine the dry heat resistance of bacterial spores exposed to heat while under vacuum or at atmospheric (760 torr) pressure. One of the few previous research efforts that specifically studied the difference in dry heat resistance of bacterial spores in a vacuum and at atmospheric pressure was the reported work of Davis, Silverman, and Keller (Ref. 1). They reported better survival of *Bacillus subtilis* var. *niger* spores at atmospheric pressure than in a vacuum of 10^{-8} to 10^{-10} torr at both 60 and 90°C.

If a true vacuum heat synergism exists which lowers the dry heat resistance of bacterial spores, then this technique, or procedure, could be utilized to sterilize spacecraft components that are heat labile. An additional advantage of this procedure would be that a lower temperature for sterilization could increase the reliability of components.

4.3.2 Approach

The initial approach was based upon the belief that the usual characterization of the thermal death of microorganisms in terms of length of exposure to a particular temperature required modification. It was proposed that the total thermal flux incident on the specimen must be considered to properly understand thermal death. The hypothesis is that the internal temperature of an organism, rather than the temperature of the surface on which it is placed, is the correct parameter. For this reason two chambers were utilized so that identical substrate temperatures with different total thermal fluxes could be studied.

A single organism, spores of *Bacillus subtilis* var. *niger*, was selected as the test organism for two reasons: (1) there was abundant dry heat survival data available in the open literature for this organism, and (2) utilization of a single test organism permitted studying a greater variety of experimental conditions within the constraint of available personnel. Additional experimental factors investigated during the initial phase of the program included: substrate, or test fixture temperatures of 80 and 100°C; chamber

wall temperatures of -185, -35, and +25° C; stainless steel and nonreflective black chamber walls (different infrared reflectivities); exposure durations of 2, 4, 6, 16, 22, 24, and 48 hr; and pressures of 10^{-5} torr and atmospheric pressure (760 torr).

4.3.3 Significant Accomplishments

4.3.3.1 Experimental Procedures. Spores of the test organism, Bacillus subtilis var. niger were sporulated in the liquid synthetic medium of Lazzarini and Santangelo (Ref. 2) modified by the addition of 25 mg of both L-methionine and L-tryptophane to a liter of medium. The inoculated medium was placed in a shaker-incubator whose temperature was set at 37°C. Mature spores were harvested and washed (7 seven times with sterile distilled water) by centrifugation (10 min at 9750 relative centrifugal force) with final suspension in distilled water.

A micropipet was used to inoculate polished aluminum stages, or tacks, with approximately 10^5 viable spores. The inoculum was allowed to dry in a humidity and temperature controlled room (45_{-0}^{+5} % relative humidity, $21 \pm 2^\circ\text{C}$ temperature). After drying, seven stages were loaded onto a polished aluminum plate interspersed with ten thermocouples. The entire test fixture is shown as Fig. 4-C.1. Film heaters were bonded to the underside of the aluminum plates of the test fixture.

Since one of the vacuum chambers was not located adjacent to the microbiology laboratory, it was necessary to transport the fixture plate under controlled conditions. This was accomplished by placing the fixture into a sealable case with inlet and outlet vales. The fixture plate was then covered with sterile aluminum foil and the box sealed and purged with extra high purity dry nitrogen.

After a test exposure, the fixture was returned to the microbiological laboratory with the inoculated stages being assayed in a rapid, consistent manner. The stages were removed from the fixture, placed individually into tubes containing 10 ml of 0.1% sterile peptone water and insonated for 12 minutes at 25 KHz in an ultrasonic bath. Upon removal from the ultrasonic bath the

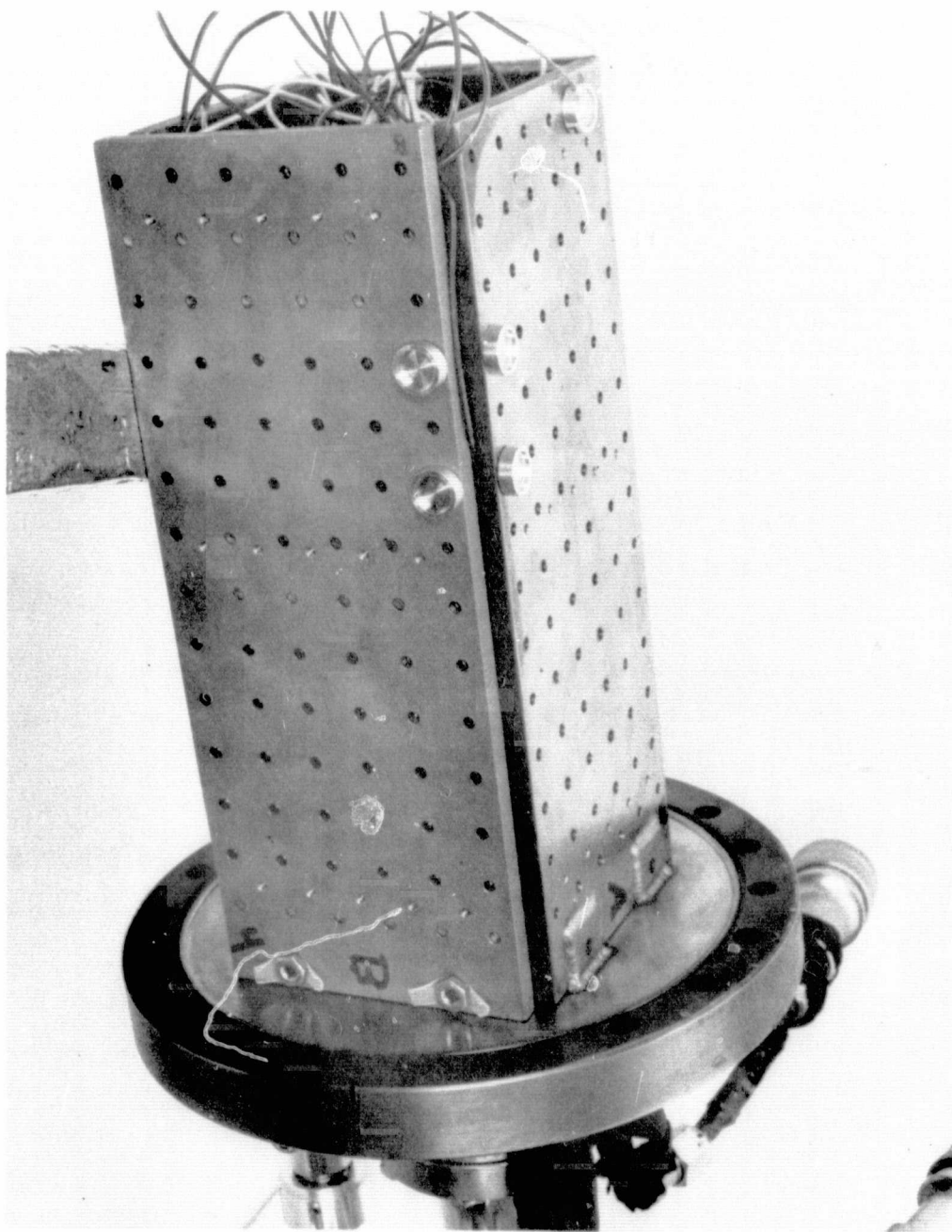


Fig. 4-C.1. Test fixture

tubed suspensions were thoroughly mixed by vortexing prior to performing serial 10-fold dilutions. Triplicate 1.0 ml aliquots of the serial dilutions were plated by the pour plate technique using trypticase Soy Agar (TSA:BBL) as the recovery medium. Plates yielding 30 to 300 colony forming units after incubation at 37°C for 48 hrs were enumerated. Survival fractions were computed by ratioing bacterial populations, recovered from seven stages after exposure to the test conditions to bacterial populations recovered from seven stages not exposed to the test conditions.

4.3.3.2 Results and Discussion. Figure 4-C.2 shows the effect of pressure on the dry heat resistance of B. subtilis spores in the stainless steel walled chamber. The chamber wall was 25°C with a substrate, or fixture plate, temperature of 80°C. Exposure durations with both sets of test conditions were 2, 4, 6, 16, and 22 hrs. Survival of the test organism was significantly higher when the chamber pressure was maintained at atmospheric pressure compared to survival fractions when chamber pressure was maintained at 10^{-5} torr pressure. Another factor requiring additional investigation was the effect of flushing the chamber with sterile GN_2 prior to establishing the vacuum. The tests at atmospheric pressure did not include a sterile GN_2 flush. Tests of this nature are planned as part of the future activities of this program.

The effect of chamber wall reflectivity and chamber wall temperature on the survival of B. subtilis spores is shown in Fig. 4-C.3. The reciprocals of the slopes of the survivor curves, in terms of hours required to lower the population one log, are given to emphasize the distinctiveness of the different test conditions and are not presented necessarily as D value equivalents. Survival in a vacuum and a substrate temperature of 80°C was best in the stainless steel walled chamber with a wall temperature of -35°C and least in this same chamber with a wall temperature of +25°C.

The effect of chamber wall reflectivity and chamber wall temperature at a substrate temperature of 100°C on the survival of B. subtilis spores is shown in Fig. 4-C.4. The ordering of reciprocals of the slopes is similar to those occurring at 80°C but the closer distribution of the values found with 100°C indicate that as the temperature of the substrate is increased the effects from wall reflectivity and wall temperature are lessened.

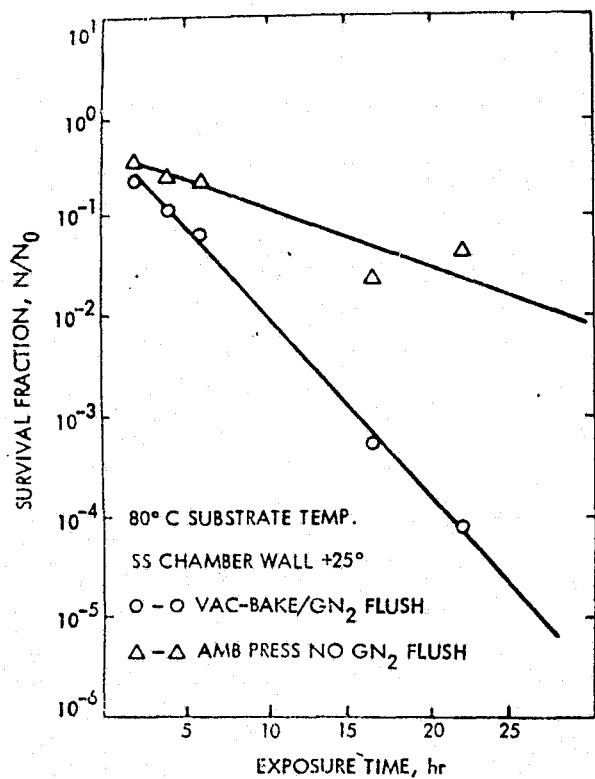


Fig. 4-C.2. Effect of pressure on dry heat resistance of Bacillus subtilis

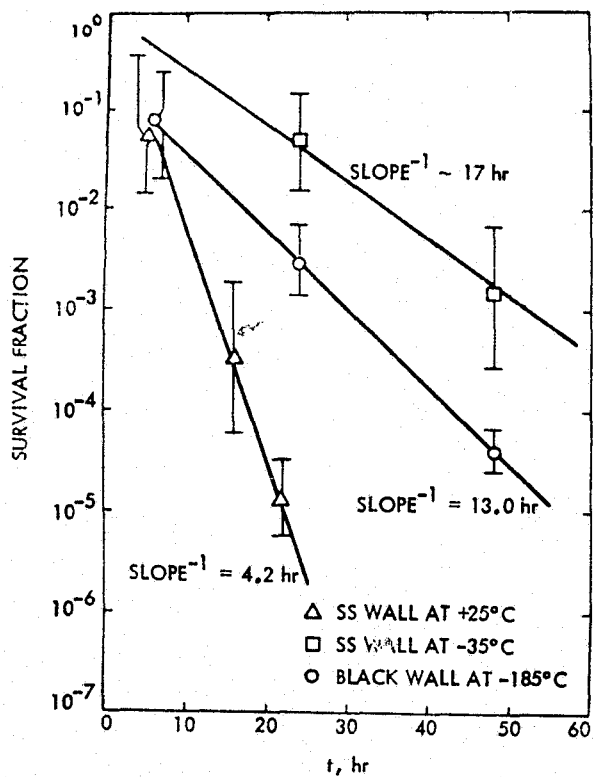


Fig. 4-C.3. BSN in vacuum
 $T_o = 80^\circ \text{C}$

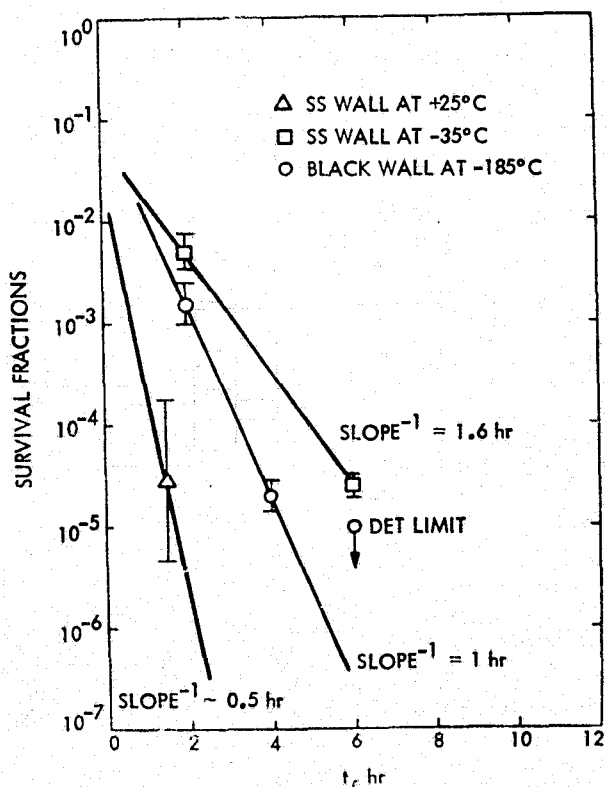


Fig. 4-C.4. BSN in vacuum
 $T_o = 100^\circ \text{C}$

The observed effect of chamber wall infrared reflectivity and temperature on the thermal death of spores placed on a substrate at constant temperature may be interpreted in terms of the internal temperature of the spores. In general the equilibrium temperature of a specimen is determined by a thermal balance equation, where the algebraic sum of all thermal inputs is set to zero.

The application of this principle to the present results may be illustrated with the aid of Fig. 4-C.5. A major term in the thermal balance equation is the conduction from the tack to the spore, which is proportional to the temperature difference $T_o - T_B$. In the absence of all other thermal transport terms, conduction would drive $T_o = T_B$, the usual simple result. Other thermal flux modes include, however, thermal emission from the substrate which is reflected from the outer wall and thermal emission from the outer wall, part of which is absorbed by the spore. These terms are always positive, tending to drive $T_o < T_B$. Finally there is thermal emission from the spore itself, which is negative and tends toward $T_o > T_B$.

The conduction may be parameterized uniquely by an effective conductivity. The conductivity depends on spore physiology and possibly deposition method and substrate surface parameters. The thermal emission from the spore depends on its infrared absorptivity to emissivity ratio (α/ϵ) which is typically near unity. Neither of these factors are convenient experimental parameters. Reflected and direct thermal emission from the outer wall may be easily varied in a known way by changes in outer wall temperature and infrared reflectivity. These terms also depend on substrate temperature, substrate reflectivity, spore absorptivity, and geometry, however.

Preliminary calculations indicate that in certain circumstances spore/substrate temperature differences of as much as 10°C are possible. The analysis is quite imprecise at present because of inconvenient geometry and the difficulty in normalizing data from two different vacuum systems. The effective conductivity of the spore/substrate interface is also only a crude estimate. Two predictions have been experimentally verified, however. Outer wall temperature and reflectivity do affect thermal death. This effect decreases as substrate temperature increases, and conduction tends to dominate.

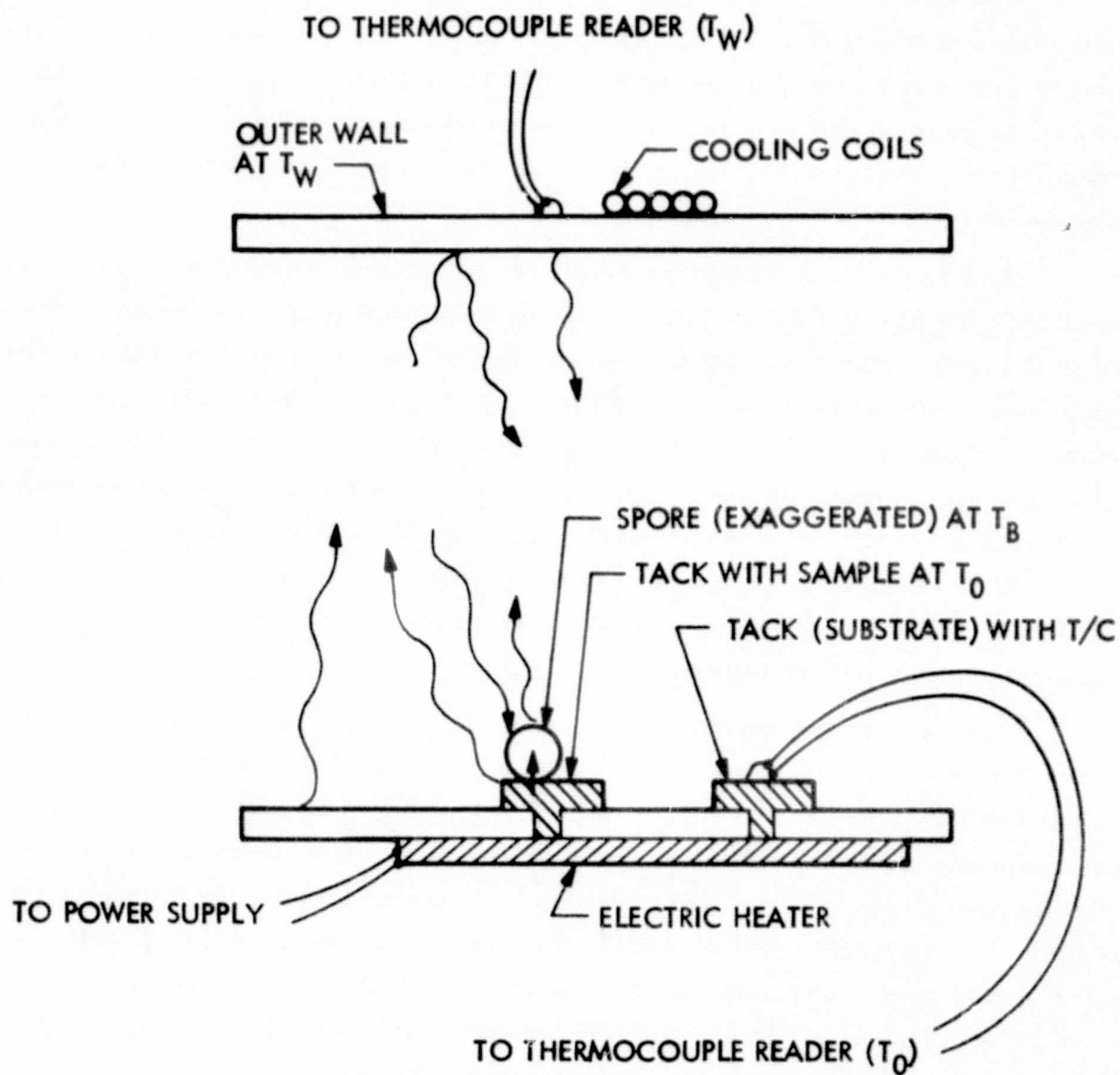


Fig. 4-C. 5. Vacuum experiment schematic showing thermal flux components

4.3.4 Future Activities

Future activities of this program will attempt to elucidate and define the effects of chamber wall temperature, infrared reflectivity, and other parameters on the sterilizing efficiency of dry heat concurrently applied with vacuum and atmospheric pressure. Studies will be conducted in a unique system with good controls and a geometry which lends itself to analysis. The analysis will quantify the various components of thermal flux and the microbial internal temperature, and allow the thermal inactivation data obtained to be modeled.

4.3.5 Presentations

Barengoltz, J. B., "Evaluation of Vacuum/Heat Sterilization," presented at the NASA Spacecraft Sterilization Technology Seminar, Cocoa Beach, Florida, August, 1975.

4.3.6 References

1. Davis, N. S., Silverman, G. J., and Keller, W. H., "Combined Effects of Ultrahigh Vacuum and Temperature on the Viability of Some Spores and Soil Organisms," *Appl. Microbiol.* 11: 202-210 (1963).
2. Lazzarini, R. A. and Santangelo, E., "Medium-dependent Alteration of Lysine Transfer Ribonucleic Acid in Sporulating Bacillus subtilis Cells," *J. Bacteriol.* 94: 125-130 (1967).

4.4 CLEAN TRANSFER SYSTEMS

4.4.1 Subtask D Introduction

The objective of this task is to develop a cleanable and sterilizable seal configuration to be used as a required part of a clean or sterile transfer system. The transfer system will be required for repair of sterile spacecraft prior to launch; on board a shuttle craft; for sterile transfer of equipment or materials from one facility to another or for the placing of sample return containers or their contents into a biobarrier system. Such a transfer system must prevent the microbial contamination of materials or biobarrier systems during the transfer process. When two enclosures are mated, viable organisms can be trapped between the mated surfaces and can be transferred into the sterile barrier when the separation is made between the two surfaces. In this study a concept is being investigated which utilizes a multiple use, heat sealable, heat sterilizable seal system for the enclosure interfaces that will prevent the transfer of organisms across this barrier-to-barrier interface.

4.4.2 Approach

In order to fulfill the objectives of this task a test program was initiated to develop an understanding of the material, design and sterilization requirements of a sterilizable seal system. Seal materials were initially evaluated for their physical, chemical and biological properties at temperatures ranging from 160 to 200°C. These temperatures were chosen as a result of previous data which indicated a 5 log spore reduction within a reasonable time span of 1 to 1.5 minute.

It was decided that candidate thermal seal materials must be able to adhere strongly and uniformly over the surface of another material which will function as a heat source. Upon heating, the seal material must soften or melt to a specified depth along the interface at approximately the specified temperature, such that de-adhesion will occur and the seal material can be clearly separated from the heated surface. When the seal material is returned to press against the heated surface it must have the capability of reestablishing a strong and uniform adhesion when the system is cooled. Alternately it is

assumed that a screen or grid of heater wires can be embedded within the seal material. Upon heating the wires electrically, the seal material will soften or melt along the plane of the heater wires, such that a clean separation can be achieved.

4.4.3 Significant Accomplishments

Several materials having the properties required of the seal system have been initially evaluated, and to date the following two general classes of materials have been identified as probable candidates:

- 1) High temperature melting point crystalline waxes.
- 2) Low molecular weight polymers having high temperature melting or softening points.

A fixture was fabricated and is presently being used in the evaluation of heat seal and cutting mechanisms for use with the candidate materials in order to determine their adhesion and melting characteristics.

4.4.4 Future Activities

Work will be continued to evaluate heat seal and cutting mechanisms using a prototype fixture. After the physical properties of the most promising transfer materials have been evaluated, a working model of a transfer fixture will be fabricated and physical and microbiological tests conducted.

4.4.5 Presentation

None

4.5 ADVANCED BIO-DETECTION METHODS

4.5.1 Subtask E Introduction

The objectives of this task are to develop more efficient methods for the detection and enumeration of microbes on surfaces. Efforts will be addressed to developing techniques which require no intimate contact, are direct, provide a permanent record and produces real-time quantitative data. Such methods will fulfill the planetary quarantine requirements and eliminate pitfalls incurred in the conventional assays.

4.5.2 Approach

This study is designed to explore the applicability of certain ramifications of present technology to the development of more efficient bio-detection assays. If such technology allows remote detection of biosystems on a macroscopic scale, i. e., multispectral sensing of agricultural and forest crops as well as marine populations, perhaps this same technology could be applied to microscopy and allow the detection of microscopic biosystems such as microorganisms on a surface.

The development of a contamination assay by macrophotographic image processing will utilize macrophotography and multispectral analysis. The suitability of using lasers in bio-detection will also be pursued.

Preliminary activities involved the formulation and testing of macrophotographic specifications (films, light source, filters, exposure time and camera set-up). Special emphases were put on examining the limitations of this system relative to the smallest size object that could be adequately photographed.

Three specimen, Bacillus subtilis var. niger (spores and vegetative), glass beads (5-10 microns) and Arizona dust (5-10 microns) were used in the initial testing. These specimen were deposited on coated aluminized glass slides scribed to provide a gridwork of 4 x 6 mm (approx.) rectangles.

Macrophotographs were made of each slide using a fabricated camera set-up (JPL Photolab) Fig. 4-E.1, consisting of the following components: Polaroid MP-3 copy stand (1), Aristo light box (2), Nikon 35 mm SLR camera with long bellows extension and $f/3.5$ macro lens mounted in reverse position for a 5x photomagnification factor (3). Kodak 5069 35 mm hi-contrast copy B/W film was used for color separation photographs. Wratten filters used were: neutral 1.9 ND, blue 1.0 ND + 47B, green 1.3 ND + 55, and red 0 ND + 29. A color reference picture of each slide was taken using Kodacolor II 35 mm color film.

Photomicrographs (100x and 500x) of each specimen were taken using a Leitz microscope. The photomicrographs are to be used as baseline information.

Data processing will consist of scanning and multispectral analysis. Each B/W 35 mm color separation film frame for a given specimen will be scanned on a Perkin-Elmer PDS microdensitometer, digitized and recorded on magnetic tape.

Each set of digital color separation images will be analyzed on the GE Image-100 (I 100) system Fig. 4-E.2, to determine the existence and degree of a multispectral signature associated with any of the specimen.

4.5.3 Significant Accomplishments

During this feasibility period preliminary photographic specifications were tested, and the data processing, scanning and multispectral analysis, is now in progress.

4.5.3.1 Macrophotography. With reference to the first objective to determine the size of the smallest objects that can be detected using macrophotography, it has been demonstrated that objects as small as 2 microns can be detected using a camera and macro lens stopped to $f/8$. At this setting, however, the macrophotographic film image of any object less than about 12 microns in diameter appears as an Airy disk of 12 micron diameter due to the diffraction limit of the optics. At $f/3.5$ (wide open) the Airy size limitation should be reduced to about 6 microns. The effect this has on feature class area percentages will be evaluated.

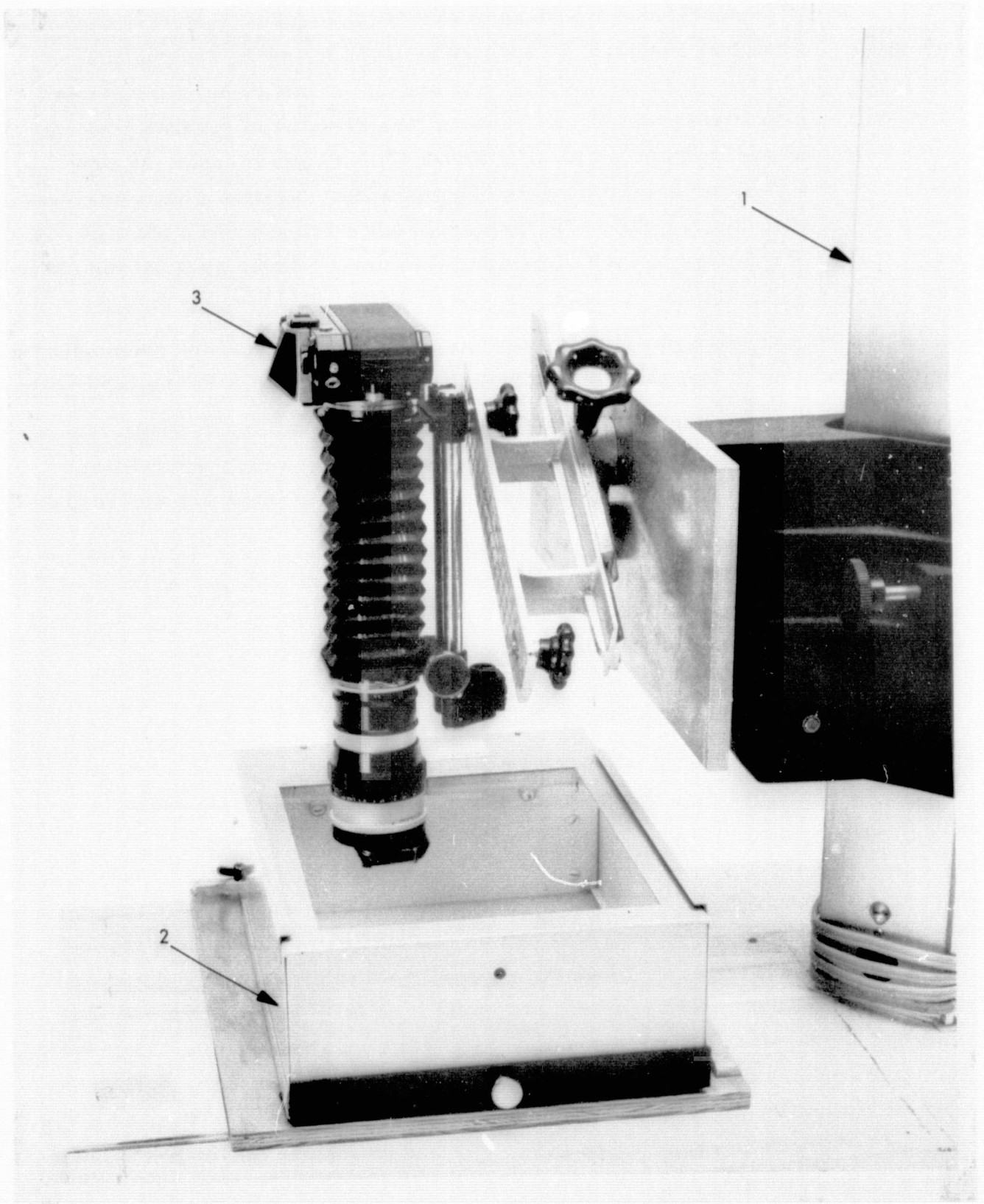


Fig. 4-E.1. Bio-detection macrophotographic camera jig

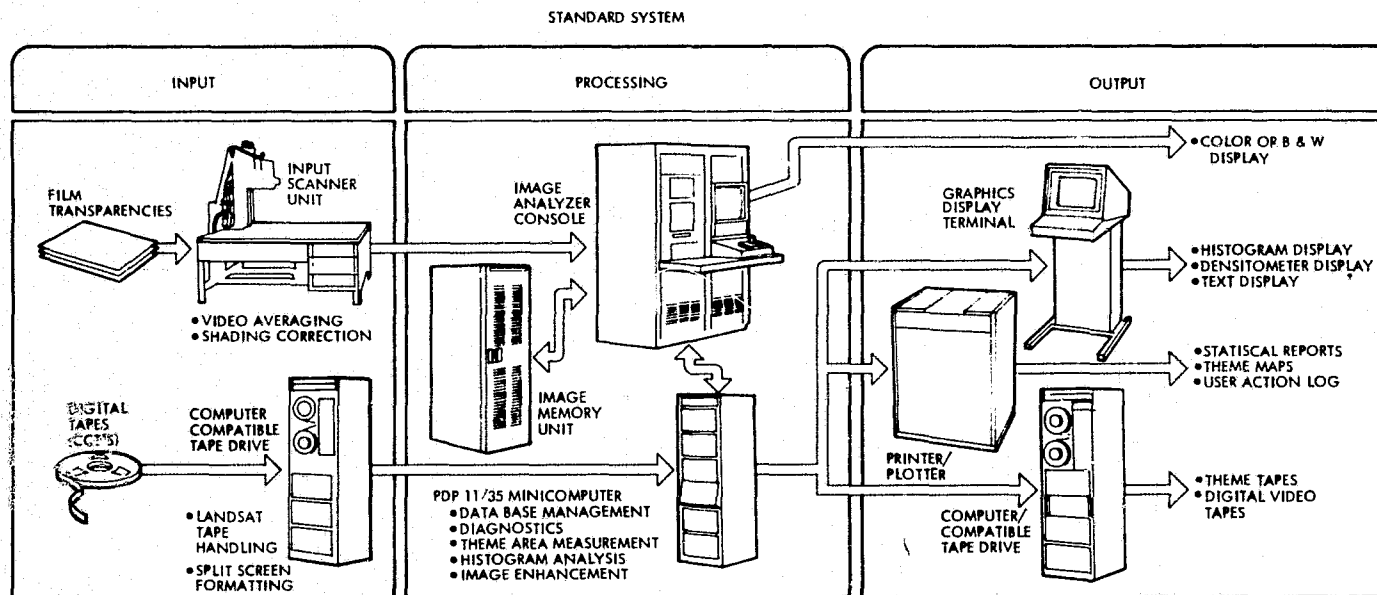


Fig. 4-E.2. Image-100 system configuration

4.5.4 Future Activities

When the results of the multispectral analysis becomes available, if there are indications of characterizing signatures for the test specimen, then experimentation will proceed. Immediate attention will be focused on the characterization of a variety of organisms and particulates.

Ultimately, a contamination assay will be developed which will determine the percent of area contaminated, and classes of contamination.

The applicability of lasers to bio-detection will be studied.

4.5.5 Presentations

None.

SECTION V

PLANETARY QUARANTINE CONSIDERATIONS
FOR
SHUTTLE LAUNCHED SPACECRAFT

Contents

para. 5.1

Title and Related Personnel

PLANETARY QUARANTINE CONSIDERA-
TIONS FOR SHUTTLE LAUNCHED
SPACECRAFT

Cognizance: M. Christensen, D. Taylor

5.1 PLANETARY QUARANTINE (PQ) CONSIDERATION FOR SHUTTLE LAUNCHED SPACECRAFT

5.1.1 Introduction

The objectives of this task are to assess the potential impact of Space Transportation System (STS) launched spacecraft (s/c) on PQ program requirements.

5.1.2 Significant Accomplishments

During the report period the present Shuttle baseline and alternate ground, launch, and on-orbit operations¹ were evaluated to determine if they could potentially impact PQ requirements for planetary missions. The primary results of this evaluation were developed under the constraints of the Shuttle Program's baseline requirements. The present program constraints are as follows:

- 1) No Spacecraft Shroud — At this stage, Shuttle planning makes no provision for a protective shroud, although some discussions are underway relative to a reusable shroud concept.
- 2) Sequential Integration — The baseline calls for a sequential s/c integration.
- 3) Shuttle Turnaround Time — Perhaps the major driver on the PQ/Shuttle assessment was the hard requirement that the Shuttle turnaround time not exceed 160 hours.

Basically, that means all s/c-orbiter integration operations must occur within 160-hours from the time the orbiter lands, culminating one mission, until it is launched on a planetary mission. This requirement automatically assumes that there will be no unexplainable interface anomalies once the s/c enters the 160-hour timeline.

For the purpose of this study, this was interpreted to mean "that if it is necessary to reprocess a s/c for PQ reasons an anomaly has occurred and the Shuttle timeline has been impacted."

¹ Mission Alternates and Operations from JPL 760-122, "STS Planetary Mission Operations Concepts Study."

5. 1. 2. 1 Ground Operations – Effect of Integration Alternatives. Figure 5-1 displays the Shuttle baseline ground operational sequence from the time the planetary s/c is released for integration with the Shuttle orbiter until lift off. It is sequential in that the operational flow is such that the s/c will be required to integrate with the major launch components in separate facilities – with the orbiter, interim upper stage (IUS) and spacecraft going to the pad as a unit. The Shuttle Program Office is presently considering two alternatives to the ground operations baseline:

- 1) Alternate one proposes taking the s/c-IUS combination directly to the pad for installation into the orbiter.
- 2) Alternate two proposes taking just the s/c to the pad for installation to the IUS/orbiter combination.

In determining the effect of the integration alternatives on the s/c, the study compared the individual alternates in terms of required facilities and number of major s/c operations needed to perform the integration (Fig. 5-2). The dotted line indicates the point at which direct control of the s/c is relinquished. Cleaning and/or sampling of the s/c for PQ reasons beyond this point would be extremely difficult.

It can be seen that the baseline operations would require the s/c to go through several facility changes and approximately 20 major integration events, 13 of which occur after control is relinquished. In contrast, Alternate 1 requires 4 facility changes and 15 major integration events, 8 of which occur after control is relinquished and, Alternate 2 only requires 3 facility changes and 13 major integration events, 5 of which occur after control is relinquished.

Based on Mariner experience, it is assumed that such facility changes, transporting of the s/c, and multiple operations significantly increase the opportunity for microbial and particulate contamination.

As such, it is concluded that of the integration options presently considered by the Shuttle Program Office, Alternate 2 would minimize the potential impact of Shuttle ground operations on PQ.

5.1.2.2 Launch/On-Orbit Operations – Effect of Shroud. Once launched, the Shuttle will use its propulsion system to come up on orbit. The orbiter payload bay doors will be opened, the payload will be deployed (the sequence of this will be varied if a shroud is used), and the s/c will be checked out, committed for the mission and released (see Fig. 5-3). The orbiter will then withdraw for a final visual check of the s/c, the IUS will be ignited and injection will occur concluding on-orbit operations (Fig. 5-4).

In the present Shuttle baseline plan, the payload will be installed directly in the cargo bay although the orbiter structural components will be covered with a flexible liner of some yet undetermined material.

Using this Shuttle "no-shroud" baseline, a contamination assessment was performed to determine what significant contamination events could occur to a PQ certified spacecraft during pad, launch, and on-orbit operations. In comparison, consideration was given to how any contamination effect could be attenuated by use of either a disposable (Mariner-type) or reuseable shroud. For the purposes of the study a "significant contamination event" was an event that subjected the s/c to the type or level of contamination that could potentially require a reprocessing of the vehicle for PQ reasons.

The results of this assessment are as follows: For the no-shroud baseline, the number of significant contaminating events from PQ verification until IUS ignition equaled 22, for the reuseable shroud concept 20 were identified, and for the disposable shroud 8 were identified.

The conclusion from this assessment is that, in view of present Shuttle pad, launch, and on-orbit operations, shroud considerations should be considered critical in maintenance of PQ requirements.

5.1.2.3 Identification of PQ/Shuttle Impact Areas. Following examination of the ground and on-orbit operations specifically for s/c integration (facilities and events) and shroud effects, the overall Shuttle plan, Program alternatives, and payload guidelines and constraints were evaluated for potential PQ impact (Fig. 5-5). Impact being defined as, "contamination of the level and type that could necessitate the processing of a s/c for PQ reasons."

The results of this evaluation indicate that 9 additional potential impact areas have been identified: 4 in the ground operations phase and 5 during the on-orbit phase. Five of these impact the 160-hour integration timeline; 4 are independent of it.

5.1.2.4 Planetary Mission Sensitivity to Identified PQ/Shuttle Impact Areas.

It was planned to examine near-term Shuttle Planetary mission operations individually to determine if the identified PQ impact areas applied, or if Mission specific operations resulted in additional impact areas. To this end the NASA Mission Model was updated with the latest inputs (e. g., Cameron-Pettingill Half-Wedge Model, Science Advisory Committee deliberations). It was concluded that the present Model was still in a state of flux, with discussions continuing on outer planet exploration as well as possible revisions assuming a positive Viking experiment.

Based on this review, it was decided to evaluate the potential STS/PQ impact areas by mission type instead of a specific mission set. The rationale being that although planetary goals might differ, mission phases relative to the STS would remain basically the same. It was assumed that the missions considered were to planets of biological interest. On this basis, the mission types were categorized as follows:

- 1) Minimal PQ impact – flybys and orbiters
- 2) Nominal PQ impact – landers, penetrators, probes
- 3) Major PQ impact – surface sample return

The identified STS/PQ impact areas were then examined according to these mission types to see if some mission types were more sensitive to STS mission phases (Fig. 6). The results confirm what might be expected – that those missions with built-in protective requirements (in terms of PQ) are less affected by potential STS operational sources.

5.1.2.5 Task Conclusions. Based on the assessment of present Shuttle Program Office plans and alternatives to date, the following conclusions can be stated:

- 1) Under present Shuttle Program baseline constraints several Shuttle operations and proposed alternatives could have a serious impact on PQ.
- 2) Integration of the S/C on the launch pad significantly reduces possible events that could result in PQ violations.
- 3) Shroud considerations are critical in maintenance of PQ requirements.
- 4) Minimal PQ missions (flybys, orbiters) may be subjected to more contamination violations than missions with built in protective requirements (e.g., bioshield). However, the proposed Shuttle operations will impact all PQ mission types.
- 5) With 160-hour turnaround allocation, present PQ assay techniques may be unsatisfactory, in terms of time requirements.

5.1.3 Future Activities

Efforts will continue in the area of reviewing Shuttle Program Office plans and alternatives to determine if additional areas of potential impact exist. Other activities will focus on defining the nature and degree of the presently identified STS/PQ impact areas. Where possible, alternatives will be suggested on how such impact might be minimized. In addition, PQ monitoring and verification procedures will be evaluated to determine if they are adequate for application during the STS era.

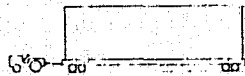
5.1.4 Presentations

Christensen, M., "Planetary Quarantine Considerations for Shuttle Launched Spacecraft," presented at NASA Spacecraft Sterilization Technology Seminar, Cocoa Beach, Florida, August 1975.

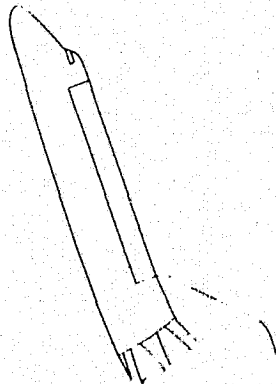
Sequential Integration



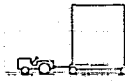
**SPACECRAFT ASSEMBLY AND
CHECKOUT IN EXPLOSIVE SAFE
FACILITY (WITHOUT RTG's)**



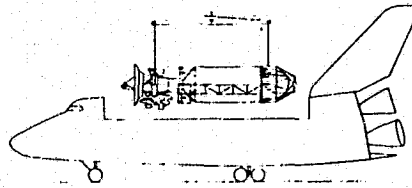
**TRANSPORT TO OPF
(HORIZONTAL TRANSPORT)**



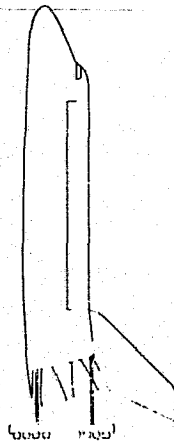
ERECT ORBITER (VAB)



TRANSPORT S/C TO SAEF



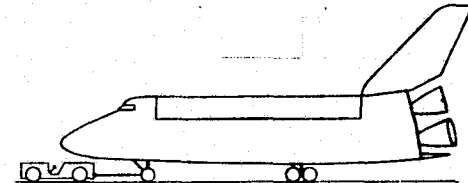
**MATE PAYLOAD TO
ORBITER IN OPF**



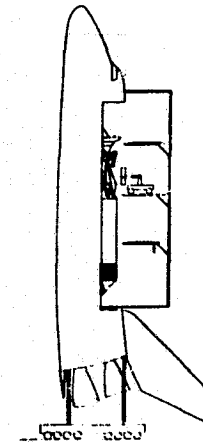
MOVE TO PAD



**INTEGRATE S/C WITH
IUS IN SAEF**



TOW TO VAB



**INSTALL RTG's, FINAL
CHECKOUT AND LAUNCH**

Fig. 5-1. Shuttle ground operational flow (baseline)

| OPERATIONAL PLANS | FACILITIES | | | | | | | MAJOR S/C OPERATIONAL EVENTS REQUIRED |
|--|--------------|-----|------|-----|-----|-----|-------|---|
| | HANGAR AO | ESF | SAEF | OPF | VAB | PAD | TOTAL | |
| STS PROJECT BASELINE (SEQUENTIAL INTEGRATION) | X | X | X | X | X | X | 6 | 20/13 |
| ALT 1 (ON-PAD) SPACECRAFT/INTERIM UPPER STAGE COMBINATION INTEGRATED WITH ORBITER | X | X | X | | | X | 4 | 15/8 |
| ALT 2 (ON-PAD) SPACECRAFT MATED TO INTERIM UPPER STAGE ORBITER COMBINATION | X | X | | | | X | 3 | 13/5 |

Fig. 5-2. Effect of integration alternatives on required facilities and major S/C operational events

SEQUENCE (BASELINE)

- ACQUIRE PARKING ORBIT
- OPEN ORBITER BAY DOORS
- DEPLOY
(VARIATION BASED ON SHROUD)
- COMMIT PAYLOAD

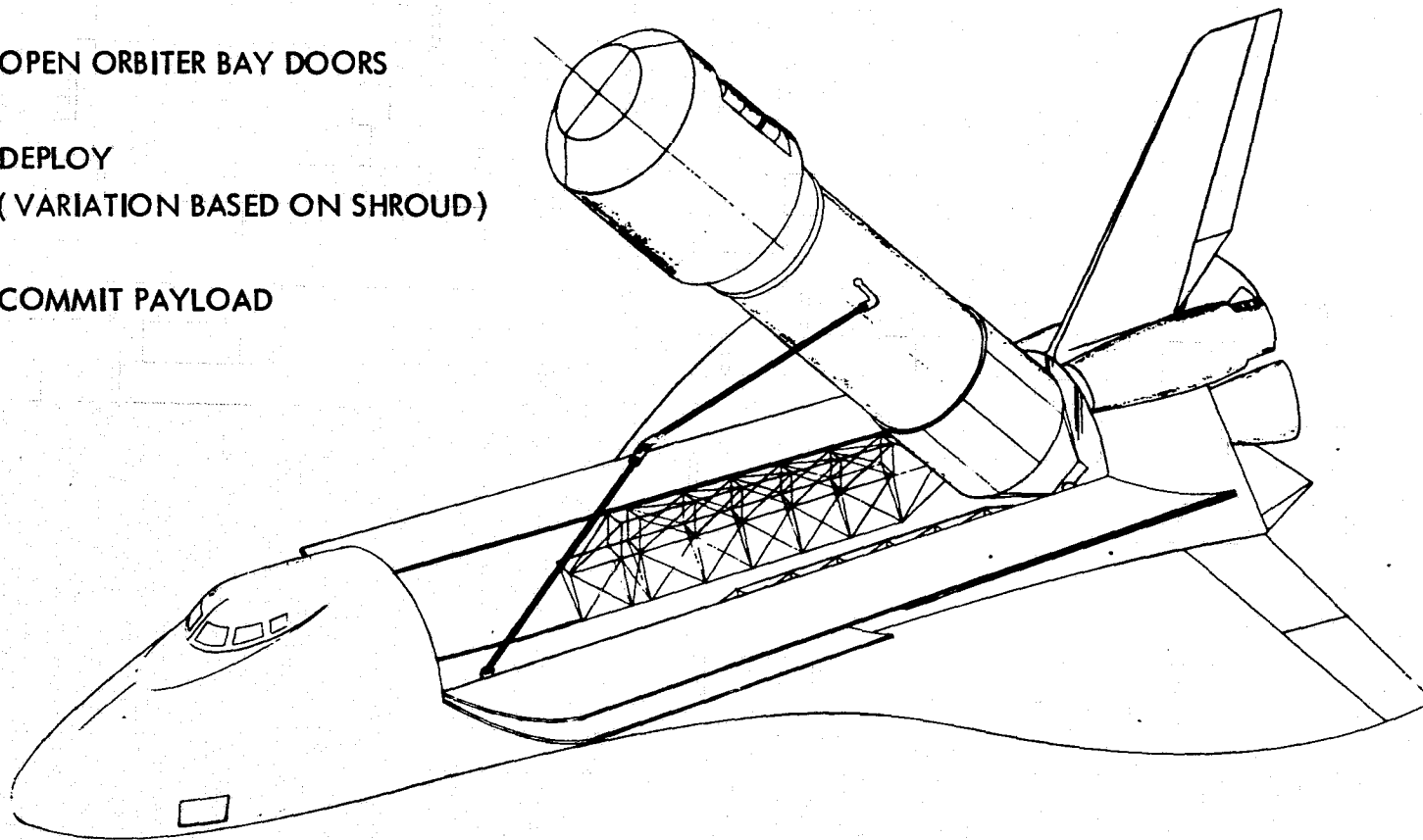
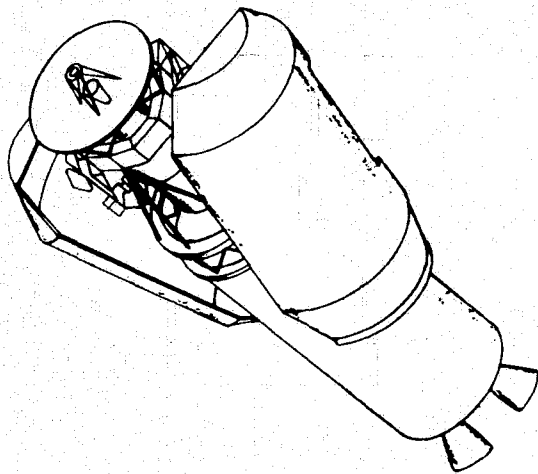


Fig. 5-3. Shuttle launch and on-orbit operations



SEQUENCE (BASELINE) (Contd)

- ORBITER WITHDRAWAL
- IUS IGNITION
- INJECTION

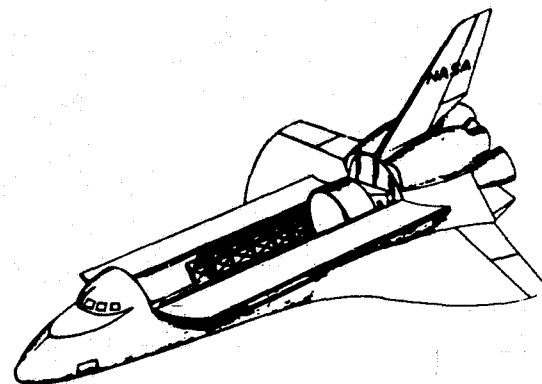


Fig. 5-4. Shuttle launch and on-orbit operations

| OPERATIONS | POTENTIAL IMPACT AREA* | IMPACT STS 160 hr TURNAROUND | |
|------------------------|--|---------------------------------|----|
| | | YES | NO |
| GROUND OPERATIONS | 1. ENVIRONMENTAL CONTROL (OPF THRU VAB) | X | |
| | 2. RTG INSTALLATION | X | |
| | 3. S/C PAD FAILURE CHANGEOUT OPTIONS | X | |
| | 4. LAUNCH COMPLEMENT (ORBITER, MLP, PAD) ASSIGNMENT | X | |
| ON-ORBIT OPERATIONS | 5. ON-ORBIT DWELL TIME | | X |
| | 6. S/C DEPLOYMENT | | X |
| | 7. ON-ORBIT REPAIR OPTION | | X |
| | 8. ABORT/RELAUNCH TIME STRATEGY | X | |
| | 9. LAUNCH AND ON-ORBIT RECONTAMINATION PROFILE | | X |

* Mission alternatives and operations from JPL 760-122, "STS Planetary Mission Operations Concepts Study."

Fig. 5-5. Identification of PQ/shuttle impact areas

| OPERATIONS | POTENTIAL IMPACT AREA | PQ MISSION TYPE | | |
|---------------------|--|-----------------|---------|------------------------|
| | | MINIMUM | NOMINAL | MAXIMUM ⁽¹⁾ |
| GROUND OPERATIONS | 1. ENVIRONMENTAL CONTROL (OPF THRU VAB) | X | | |
| | 2. RTG INSTALLATION (ON-PAD IF REQUIRED) | X | (2) | (2) |
| | 3. S/C PAD FAILURE CHANGEOUT OPTIONS | X | X | X |
| | 4. LAUNCH COMPLEMENT ASSIGNMENT | X | X | X |
| ON-ORBIT OPERATIONS | 5. ON-ORBIT DWELL TIME | X | | |
| | 6. S/C DEPLOYMENT | X | | |
| | 7. ON-ORBIT REPAIR OPTION | X | X | X |
| | 8. ABORT-RELAUNCH TIME STRATEGY | X | X | X |
| | 9. LAUNCH AND ON-ORBIT RECONTAMINATION PROFILE | X | X | X |
| | TOTAL | 9 | 5 | 5 |

¹ Return leg of SSR not considered

² Assumes any on-pad RTG installation would be external to bioshield, hence no effect.

Fig. 5-6. Planetary mission sensitivity to identified PQ/shuttle impact areas

SECTION VI

PLANETARY QUARANTINE LABORATORY ASSAY
ACTIVITIES (AFETR)
NASA No. 195-58-63-05

Contents

para. 6.1

Title and Related Personnel

PLANETARY QUARANTINE LABORATORY
ASSAY ACTIVITIES (AFETR)

Cognizance: J. R. Puleo

Associate
Personnel:

G. Oxborrow
N. Fields
S. Bergstrom
L. Maull
K. Gantner (Bionetics)
G. Hays (Bionetics)

6.1 PLANETARY QUARANTINE LABORATORY ASSAY ACTIVITIES (AFETR)

6.1.1 Introduction

The objective of this subtask is to determine and document the quantitative and qualitative microbiological profiles of pertinent automated out-bound spacecraft which may carry terrestrial organisms to the planets.

This type of work is required by NASA policy and lays a foundation upon which future policy can be formulated.

6.1.2 Significant Accomplishments

Bioassays were done on the Viking Precursor spacecraft which consisted of Viking Lander Capsule (VLC-1), Orbiter (VO-1) and Shroud B. Microbiological assessments were initiated on the Viking lander (VLC-2) and Orbiter (VO-2). Microbiological samples were taken at pre-determined milestone events by methods described in the Viking '75 Program Microbiological Assay and Monitoring Plan. Each milestone consisted of 250 swab samples plus 25 negative controls. Samples were taken from the spacecraft and assayed by the responsible organization and the Planetary Quarantine Laboratory (PQL). The PQL was responsible for assaying 50% of all samples taken as described in the Viking '75 Plan.

All bioassays were done in the three separate laboratory areas located in the PQL which were established and equipped to support the Viking Microbiological assay activities. These laboratories housed the bioassay teams from Martin-Marietta Aerospace (MMA), JPL Viking Orbiter (JPL-VO) and Planetary Quarantine Laboratory (PQL).

Bioassay teams were formed to process the microbial samples taken from the Viking spacecraft. These bioassay teams consisted of two people, one person from the responsible organization, the other, a technician supplied by the PQL. The PQL has two bioassay teams, one team works only on the Lander samples, the other team works on the Orbiter samples. This allows for simultaneous processing of samples from the Lander and Orbiter spacecraft. Members of the bioassay teams will remain together for the entire Viking Program.

Bacterial colonies were picked from culture plates at each milestone assay. Plates containing colonies were saved and colonies were randomly picked. Table 6-1 shows the number of colonies isolated to date. These isolates will be identified, lyophilized and stored.

The quantitative results obtained from the bioassays were submitted to the Planetary Quarantine Officer and Viking Project Office. As the qualitative results become available, they will also be sent to the Planetary Quarantine Officer.

A new cooperative study was initiated with Hardin-Simmons University. The purpose of this study was to obtain a more complete microbiological characterization of organisms associated with the Viking spacecraft. Specifically, the study was to determine the presence of mesophilic and psychrophilic anaerobes on the Viking Spacecraft. The PQL provided laboratory space to personnel from Hardin-Simmons University to conduct this investigation.

Table 6-1. Number of Microorganisms Isolated For Identification From Viking Spacecraft

| | | NHS ^a | HS ^b |
|---|---------------|------------------|-----------------|
| <u>PRECURSOR</u> | VLC-1 | 51 | 73 |
| | VO-1 | 128 | 5 |
| | SHROUD B | 32 | 2 |
| <u>VIKING A</u> | VLC-2 | | |
| | Milestone C-3 | 0 | 21 |
| | Milestone C-4 | 0 | 53 |
| | VO-2 | | |
| | Milestone O-3 | 76 | 24 |
| | Milestone O-4 | 38 | 3 |
| | Solar Panels | 12 | 4 |
| | Total | 337 | 185 |
| ^a Vegetative organisms; non-heat shocked | | | |
| ^b Spore formers; heat shocked | | | |

6.1.3 Future Activities

Complete the bioassay of the Viking Lander Capsule and Orbiter spacecraft. Bacterial colonies resulting from the Viking bioassays will be picked randomly from culture plates, gram stained and identified. These isolates will be maintained on the appropriate media, subsequently lyophilized and stored for future reference. The quantitative and qualitative microbial assessment of the Viking Lander Capsule and Orbiter spacecraft will be submitted to the Planetary Quarantine Officer and Viking Project Office. Microbiological profiles will be conducted on pertinent spacecraft as requested by the Planetary Quarantine Officer.

6.1.4 Presentations

Puleo, J. R., Status of Viking Planetary Quarantine Operations, AIBS Planetary Quarantine Panel, April 1975, Cocoa Beach, Florida.

6.1.5 Publications

Oxborrow, G. S., N. D. Fields, J. R. Puleo, and C. M. Herring, "Quantitative Relationship between Airborne Viable and Total Particles", Health Lab Sci., Vol. 12, No. 1, Jan. 1975.

SECTION VII

PLANETARY QUARANTINE LABORATORY - RESEARCH
ACTIVITIES
(NASA No. 193-58-63-06)ContentsSubtask A
para. 7.1Title and Related Personnel

TEFLON RIBBON EXPERIMENTS

Cognizance: J. R. Puleo

Associate
Personnel: G. Oxborrow
N. Fields
S. BergstromSubtask B
para. 7.2PYROLYSIS GAS-LIQUID
CHROMATOGRAPHY STUDY

Cognizance: G. Oxborrow

Associate
Personnel: N. Fields
J. R. Puleo

7.1 TEFLON RIBBON EXPERIMENTS

7.1.1 Subtask A Introduction

The objective of this study is to characterize the thermal resistance profiles of naturally occurring bacterial spores associated with assembly facilities at Kennedy Space Center.

The validity of the currently accepted sterilization cycle should be confirmed. The cycle, to be valid, should be effective on bacterial spores associated with spacecraft in residence at Kennedy Space Center, Florida.

7.1.2 Significant Accomplishments

7.1.2.1 Results. The teflon ribbon study was continued. The details of the thermal apparatus and experimental test procedures were described in para. 5.1.2.1 of Jet Propulsion Laboratory (JPL) Doc. No. 900-655, April 1974. Naturally occurring airborne bacterial spores were collected on teflon ribbons exposed to the intramural environment of the Vehicle Assembly Building (VAB), Kennedy Space Center (KSC), Florida.

In any dry heat sterilization cycle there are several factors which may effect the efficiency of the process. One of these parameters is the duration of the thermal exposure. Table 7-A.1 shows the results obtained when the moisture level is held constant and the thermal profile is varied only by controlling the time at 111.7C. The "Survivor Fraction" (para. 5.1.2.1, JPL Doc. No. 900-675, December 1974) of the "hardy" organisms is increased from a mean of 3.2×10^{-4} to 1.3×10^{-3} and 1.4×10^{-3} when the time at 111.7C is reduced from 30 hrs. to 20 and 15 hrs. respectfully. Additional experiments will be conducted at these lower time exposures.

Table 7-A.2 shows the results obtained when the teflon ribbons were subjected to a water concentration of 0.01 mg/liter and the 11.7C high thermal inertia cycle. Survivors were recovered from all 19 experiments. The mean "Survivor Fraction" for this group of experiments was 3.0×10^{-4} .

A second dry heat oven was installed adjacent to the existing oven in the vertical laminar flow clean bench. Studies were conducted to determine

Table 7-A.1. Thermal Resistance of Bacterial Spores
Collected on Teflon Ribbons - VAB-KSC

TEMP. 111.7C
1.2 mg/Liter water

| Experiment Number | N _o Spores | Positive/Total | MPN for Ribbon | Survivor Fraction N _H /N _o |
|------------------------------------|-----------------------|----------------|----------------|--|
| Total time at temperature 14 hours | | | | |
| T-15 | 5.7×10^2 | 23/24 | 3.179 | 5.6×10^{-3} |
| T-16 | 9.9×10^2 | 22/24 | 2.485 | 2.5×10^{-3} |
| T-17 | 5.6×10^2 | 12/24 | 0.693 | 1.2×10^{-3} |
| T-19 | 2.2×10^3 | 19/24 | 1.569 | 7.1×10^{-4} |
| T-21 | 1.5×10^3 | 19/24 | 1.569 | 1.0×10^{-3} |
| T-23 | 1.4×10^3 | 23/24 | 3.179 | 2.3×10^{-3} |
| <u>mean</u> | 1.2×10^3 | 118/144 | 1.712 | 1.4×10^{-3} |
| Total time at temperature 20 hours | | | | |
| T-14 | 6.0×10^2 | 15/24 | 0.981 | 1.6×10^{-3} |
| T-18 | 1.6×10^3 | 21/24 | 2.080 | 1.3×10^{-3} |
| <u>mean</u> | 1.1×10^3 | 36/44 | 1.387 | 1.3×10^{-3} |
| Total time at temperature 30 hours | | | | |
| T-12 | 1.2×10^3 | 9/24 | 0.470 | 3.9×10^{-4} |
| T-13 | 6.6×10^2 | 10/24 | 0.539 | 8.2×10^{-4} |
| T-25 | 4.2×10^3 | 14/24 | 0.876 | 2.1×10^{-4} |
| T-26 | 1.3×10^3 | 10/24 | 0.539 | 4.1×10^{-4} |
| <u>mean</u> | 1.8×10^3 | 43/96 | 0.594 | 3.2×10^{-4} |

Table 7-A.2. Thermal Resistance of Bacterial Spores
Collected on Teflon Ribbons - VAB-KSC

TEMP. 111.7C
0.01 mg/Liter water

| Experiment Number | N _o Spores | Positive/ Total | MPN for Ribbon | Survivor Fraction N _H /N _o |
|-------------------|-----------------------|--------------------|-------------------|---|
| T-27 | 1.2 x 10 ³ | 2/24 | 0.087 | 7.2 x 10 ⁻⁵ |
| T-28 | 3.7 x 10 ³ | 19/24 | 1.569 | 4.2 x 10 ⁻⁴ |
| T-29 | 2.8 x 10 ³ | 9/24 | 0.470 | 1.7 x 10 ⁻⁴ |
| T-30 | 3.6 x 10 ³ | 20/24 | 1.792 | 4.9 x 10 ⁻⁴ |
| T-31 | 2.4 x 10 ³ | 20/24 | 1.792 | 7.3 x 10 ⁻⁴ |
| T-32 | 2.1 x 10 ³ | 19/24 | 1.569 | 7.3 x 10 ⁻⁴ |
| T-33 | 4.9 x 10 ³ | 22/24 | 2.485 | 5.1 x 10 ⁻⁴ |
| T-34 | 1.7 x 10 ³ | 17/24 | 1.232 | 7.2 x 10 ⁻⁴ |
| T-35 | 3.9 x 10 ³ | 20/24 | 1.792 | 4.6 x 10 ⁻⁴ |
| T-36 | 5.3 x 10 ³ | 24/24 | — | — |
| T-37 | 5.6 x 10 ³ | 15/24 | 0.981 | 1.7 x 10 ⁻⁴ |
| T-38 | 3.2 x 10 ³ | 11/24 | 0.613 | 1.9 x 10 ⁻⁴ |
| T-39 | 7.8 x 10 ³ | 24/24 | — | — |
| T-40 | 8.5 x 10 ³ | 20/24 | 1.792 | 2.1 x 10 ⁻⁴ |
| T-41 | 3.4 x 10 ³ | 23/24 | 3.179 | 9.4 x 10 ⁻⁴ |
| T-42 | 5.7 x 10 ³ | 14/24 | 0.876 | 1.5 x 10 ⁻⁴ |
| T-43 | 4.8 x 10 ³ | 15/24 | 0.981 | 2.1 x 10 ⁻⁴ |
| T-44 | 3.1 x 10 ³ | 11/24 | 0.613 | 1.9 x 10 ⁻⁴ |
| T-45 | 2.8 x 10 ³ | 13/24 | 0.780 | 2.8 x 10 ⁻⁴ |
| <u>mean</u> | 4.0 x 10 ³ | 318/456 | 1.195 | 3.0 x 10 ⁻⁴ |

if both ovens were compatible. Oven characteristics were found to be identical after both ovens were challenged with teflon ribbons, soil, and spore suspensions.

Ninety-eight heat survivors were randomly selected from the total survivors recovered when teflon ribbons were subjected to a water concentration of 0.01 mg/liter and the 111.7C high thermal inertia cycle (table 7-A.2). From these survivors, 73 isolates have been identified to date. Table 7-A.3 shows that *Bacillus lentus* was recovered most frequently with the atypical *Bacillus* occurring with the next most frequency. The majority of those organisms classified as belonging to the *B. lentus* group had only one positive biochemical test reaction, i. e. starch. Sixty-eight percent of the atypical *Bacillus* accounted for approximately 90% of the "hardy" organisms recovered. These data are consistent with those obtained in previous work (JPL Doc. No. 900-675, September 1974; JPL Doc. No. 900-701, April 1975).

Cooperative studies were done in collaboration with Dr. J. E. Campbell, Food and Drug Administration (FDA), Cincinnati, Ohio, and Dr. T. Foster, Hardin-Simmons University, Abilene, Texas. The dry heat inactivation characteristics of pure spore suspensions and heterogenous spore populations in soil from various geographic areas, were investigated. The studies consisted of subjecting the various spore containing menstra to the high thermal inertia sterilization cycle in the dry heat oven. Various parameters, such as, temperature, water concentration, time at temperature, or combinations of the pre-mentioned thermal conditions were varied.

Due to the Viking '75 Program this study was discontinued in May. It will resume after all the microbiological commitments have been satisfied.

Identifications were completed on eighteen bacterial cultures imported from the U. S. S. R. space program. These isolates had been sent to Dr. L. B. Hall, Planetary Quarantine Officer, by Professor V. I. Vashkov of Russia, in a culture exchange of organisms resistant to dry heat. Examination of the cultures showed that they belonged to the genus *Bacillus*. A listing of the identifications were submitted to the Planetary Quarantine Officer.

Table 7-A.3. Biochemical Test Reactions of Heat-Stressed Environmental Isolates - VAB-KSC

TEMP. 111.7C
0.01 mg/l water

| Organism | Number of Isolates | Mannitol | Tyrosine | Phenyl-alanine | Casein | Starch | Voges-Proskauer | Citrate | Nitrate | Anaerobic Growth |
|--------------------------|--------------------|----------|----------|----------------|--------|--------|-----------------|---------|---------|------------------|
| <u>B. circulans</u> | 5 | - | - | - | - | + | - | - | - | + |
| <u>B. circulans</u> | 2 | - | - | - | - | - | - | - | - | + |
| <u>B. lentus</u> | 36 | - | - | - | - | + | - | - | - | - |
| <u>B. lentus</u> | 3 | - | - | - | - | + | - | - | + | - |
| <u>B. lentus</u> | 1 | + | - | - | - | + | - | - | + | - |
| <u>B. lentus</u> | 3 | + | - | - | - | + | - | - | - | - |
| <u>B. subtilis</u> | 1 | + | - | - | + | + | + | - | + | - |
| Atypical <u>Bacillus</u> | 15 | - | - | - | - | - | - | - | - | - |
| Atypical <u>Bacillus</u> | 1 | - | - | - | - | - | - | - | + | + |
| Atypical <u>Bacillus</u> | 2 | - | - | - | - | + | - | - | + | + |
| Atypical <u>Bacillus</u> | 1 | - | - | - | - | - | - | - | + | - |
| Atypical <u>Bacillus</u> | 1 | + | - | - | + | + | + | - | - | - |
| Atypical <u>Bacillus</u> | 1 | + | - | - | + | + | + | - | + | - |
| Atypical <u>Bacillus</u> | 1 | - | - | - | + | + | - | - | - | - |

7.1.3 Future Activities

Future work will be directed to:

- 1) characterize the thermal resistance profiles of naturally occurring bacterial spores from assembly facilities at KSC.
- 2) develop thermal death curves using various temperatures, water concentrations and duration of thermal exposure.
- 3) develop and field evaluate assay techniques and procedures.
- 4) continue cooperative studies with Hardin-Simmons University to determine the occurrence of thermal resistance of mesophilic and psychrophilic anaerobic microorganisms.

- 5) continue cooperative studies with Food and Drug Administration to determine thermal inactivation of homogenous and heterogenous bacterial spore populations.
- 6) characterize the thermal resistant actinomycetes recovered from the teflon ribbon study, and determine their heat inactivation kinetics.
- 7) determine thermal inactivation kinetics of bacterial spore populations isolated from the Viking Lander Capsule and Orbiter spacecraft.

7.1.4 Publications

Puleo, J. R., Favero, M. S., Oxborrow, G. S., and Herring, C. M. A Method for Collecting Naturally Occurring Airborne Bacterial Spores for Determining their Thermal Resistance. Accepted for publication in the journal of Applied Microbiology on August 12, 1975, to appear in November 1975 issue.

7.2 PYROLYSIS GAS-LIQUID CHROMATOGRAPHY STUDY

7.2.1 Subtask B Introduction

The objective of this study is to develop a rapid and reliable method for the identification of microorganisms utilizing pyrolysis gas - liquid chromatography techniques and computer technology.

Dry heat resistant bacterial isolates surviving the terminal sterilization process for unmanned landers are difficult or impossible to identify by conventional morphological and biochemical methods. Isolates with identical biochemical characteristics often have different colonial, cellular and spore morphology. Pyrolysis gas - liquid chromatography is a rapid and sensitive method of bioorganic analysis and has been demonstrated to give consistent chromatographic analysis of microorganisms.

7.2.2 Significant Accomplishments

7.2.2.1 Results. Evaluation of pyrolysis gas - liquid chromatography (PGLC) as a tool for the rapid identification or characterization of micro-organisms was continued. Microbial cells grown on membrane filters were compared to agar grown cells and found to give more consistent chromatograms from replicate plates. This technique eliminated the need for washing cells to remove possible agar contamination, and also more consistent sample sizes were easier to obtain.

Microbial growth on different lot numbers of Trypticase Soy Agar (TSA;BBL) were compared to assess the chromatographic differences. Preliminary studies show little chromatographic differences were obtained when four species of *Bacillus* were grown on membrane filters using three lot numbers of TSA. However, when different media (TSA, Eugon Agar, Nutrient Agar) were compared, differences were observed.

Preliminary results indicate that bacterial cells should be harvested during the stationary or death phase for obtaining better chromatograms.

A glass column 1/8 inch x 20 feet packed with 5% carbowax 20m TPA on Anikrom ABS 100-110 mesh was compared to the previously used 1/8 inch x 10 feet stainless steel column packed with 7% carbowax 20m TPA on Anikrom ABS 100-110 mesh and was found to give much better peak separations. The glass column separated 60-64 chromatographic peaks compared to 43-47 on the stainless steel column.

Due to the Viking '75 Program this study was discontinued in March. It will resume after all the microbiological commitments have been satisfied.

7.2.3 Future Activities

Future activities will be directed to: 1) continue pyrolysis of known bacterial cultures and environmental microbial isolates to complete a chromatographic catalogue using computer analysis, 2) determine correlation between physiological characteristics and peak variation, 3) identify or

characterize dry heat resistant microorganisms, 4) assess the effect of culturing techniques, 5) evaluate a pattern recognition computer identification system, 6) chemically identify resultant chromatographic peaks using mass spectroscopy, and 7) initiate cooperative studies with Hardin-Simmons University for characterizing mesophilic and psychrophilic anaerobic microorganisms.